d 19 cit,ab 1

1. 4,738,927, Apr. 19, 1988, Gene coded for interleukin-2 polypeptide, recombinant DNA carrying the said gene, a living cell line possessing the recombinant DNA, and method for producing interleukin-2 using the said cell, Tadatsugu Taniguchi, et al., 435*243, 69.52, 91, 172.1, 172.3, 240.2, 252.31, 252.33, 255, 256, 317.1, 320, 849; 530*351; 536*27; 935*11, 69, 73

US PAT NO: 4,738,927 L9: 1 of 1

ABSTRACT:
A gene coded for a polypeptide which possesses interleukin-2 activity is isolated, and connected with a vector DNA which is capable of replicating in a procaryotic or eucaryotic cell at a position downstream of a promoter

US PAT NO: 4,738,927 L9: 1 of 1
gene in the vector obtaining a recombinant DNA, with which the cell is transformed to produce interleukin-2.

=>

d l19 cit,ab 2,4

2. 4,889,919, Dec. 26, 1989, Biologically active PDGF derived A-chain homodimers; Mark J. Murray, et al., 530*351; 435*69.4, 172.3; 514*2, 8, 21; 530*350, 380, 399

US PAT NO: 4,889,919 L19: 2 of 5

US PAT NO: 4,889,919 L19: 2 of 5

ABSTRACT:
Dimeric Proteins having substantially the same biological activity as PDGF are disclosed. More specifically, the Protein may have two substantially identical polypeptide chains, each of the chains being substantially homologous to the A-chain of PDGF. Alternatively, the Protein may have two polypeptide chains that are substantially identical to the A-chain of PDGF. In addition, Proteins comprising polypeptides that are variants or derivatives of the A-chain of PDGF are also disclosed. Therapeutic compositions containing these Proteins and methods for enhancing the wound-healing process in warm-blooded animals are also disclosed.

4. 4,766,073, Aug. 23, 1988, Expression of biologically active PDGF analogs in eucaryotic cells; Mark J. Murray, et al., 435*69.4, 91, 172.3, 255, 317.1; 935*13, 28,

US PAT NO: 4,766,073 L19: 4 of 5

ABSTRACT:
Methods for expressing a variety of biologically active PDGF analogs in eucaryotic cells are disclosed. The methods generally comprise introducing into a eucaryotic host cell a DNA construct capable of directing the expression and secretion of biologically active PDGF analogs in eucaryotic cells. The DNA construct contains a transcriptional promoter followed downstream by a suitable DNA sequence. The DNA sequence may encode a Protein substantially homologous to the A-chain or the B-chain of PDGF, or a portion thereof, or an A-B heterodimer. In addition, a portion of the DNA sequence may encode at least a portion of the A-chain, while another portion encodes at least a portion of the B-chain of PDGF. Eucaryotic cells transformed with these DNA constructs are also disclosed. Methods of promoting the growth of mammalian cells, comprising incubating the cells with a biologically active PDGF

US PAT NO: 4,766,073 L19: 4 of 5
analog expressed by a eucaryotic host cell transformed with such a DNA construct, are also disclosed.

=>

d l27 cit,ab 1-3;d his;delete history

1. 4,886,747, Dec. 12, 1989, Nucleic acid encoding TGF-.beta. and its uses; Rik M. A. Derynck, et al., 435*69.4, 69.9, 172.1, 172.3, 240.1, 240.2, 320; 536*27; 935*11, 37, 79

US PAT NO: 4,886,747 L27: 1 of 3

ABSTRACT:
Nucleic acid encoding TGF-.beta. has been isolated and cloned into vectors which are replicated in bacteria and expressed in eukaryotic cells. TFG-.beta. is recovered from transformed cultures for use in known therapeutic modalities. Nucleic acid encoding TGF-.beta. is useful in diagnosis and identification of TGF-.beta. clones.

2. 4,742,003, May 3, 1988, Human transforming growth factor; Rik M. A. Derynck, et al., 435*69.4, 172.3, 255, 320; 530*324, 350, 828; 935*13, 29, 37, 47, 48

US PAT NO: 4,742,003 L27: 2 of 3

ABSTRACT:
Methods and compositions are provided for the recombinant synthesis of the tumor growth factor-.alpha. precursor and its fragments. These are useful in therapy and diagnosis, as are antibodies raised by immunization

with the tumor growth factor-alpha precursor and its fragment.

U.S. 4,670,539, Jun. 2, 1987, Peptide growth factors derived from estrogen responsive kidney tissue; David A. Sirbasku, et al., 530*324

US PAT NO: 4,670,539 L27: 3 of 3

ABSTRACT:
Two forms of polypeptide growth factors have been purified to homogeneity

US PAT NO: 4,670,539 L27: 3 of 3
from lyophilized powder of mature ewe kidneys. From 500 g of powder 8 to 14 mg of kidney derived growth factor (KDGF) was isolated. An 18,000 fold purification was accomplished with a 4 to 7% yield by a 6 step procedure that included an initial acetic acid extraction, heating at 95.degree., Bio-Rad AG50W X8 cation exchange chromatography, 2 sequential DEAE-Sepharose CL-6B anion exchange steps at pH 5.8 and 6.2, respectively, and finally Sephadex G-50 chromatography in 0.1M acetic acid. From the Sephadex molecular sieve separation, the KDGF activity eluted in the same fractions as the single Protein peak. Polyacrylamide gel electrophoresis analysis under non-reducing and non-denaturing conditions followed by Coomassie Blue staining confirmed a single band having a M.sub.r 4,200. Molecular sieve HPLC done under acidic conditions confirmed a similar high (i.e. greater than 95%) degree of homogeneity. This preparation of KDGF was shown to be mitogenic for rat pituitary tumor cells, and hamster and rat kidney tumor cells, but not for normal rat diploid fibroblasts or low passage normal rat kidney (NRK) cells. The KDGF preparation that appeared greater than 90% homogeneous by the

US PAT NO: 4,670,539 L27: 3 of 3
criteria described above was separated by chromatofocusing into 2 forms, KDGF-I and KDGF-II, of pI 5.2 and 4.8, respectively. Both forms showed M.sub.r 4200. KDGF-II was the more abundant form, being 70% of the total KDGF isolated from the chromatofocusing elution. The G.sub.50s of KDGF-I and KDGF-Ii were 10 ng/ml (2.38.times.10.sup.-9 M) and 19 ng/ml (4.52.times.10.sup.-9 M), respectively, with the MTW9/PL cells in culture without any other additions to the serum free medium used in these bioassays.

(FILE USPAT)

DELETE HISTORY

L1	6097	S	DNA OR RNA OR NUCLEIC
L2	1021	S	MATSUI?/IN OR AARONSON?/IN
L3	1409	S	L2 OR PIERCE?/IN
L4	9606	S	RECEPTOR?
L5	799	S	GROWTH(2W)FACTOR
L6	52	S	PDGF
L7	804	S	L5 OR L6
L8	141	S	L7 AND L4 AND L1
L9	1	S	L8 AND L3
L10	140	S	L8 NOT L9
L11	769398	S	TYPE?
L12	124	S	L10 AND L11
L13	95	S	SEQUENCE? AND L12
L14	29024	S	PROTEIN?
L15	92	S	L14 AND L13
L16	7068	S	ANTIBODY
L17	64	S	L16 AND L15
L18	166	S	RECEPTOR(2W)PROTEIN
L19	5	S	L17 AND L18
L20	161	S	L18 NOT L19

```
L21           59 S L17 NOT L19
L22        67017 S HUMAN
L23           58 S L22 AND L21
L24         1595 S RECEPTOR?/AB,TI
L25            9 S L23 AND L24
L26           52 S PDGF
L27            3 S L26 AND L23


DO YOU WANT TO DELETE ALL L# ITEMS? (Y)/N:y
```

?LOG OFFt s9/7/1-14

9/7/1 (Item 1 from file: 399)
112071609 CA: 112(9)71609x JOURNAL
An efficient directional cloning system to construct cDNA libraries containing full-length inserts at high frequency
AUTHOR(S): Miki, Toru; Matsui, Toshimitsu; Heidaran, Mohammad A.; Aaronson, Stuart A.
LOCATION: Lab. Cell. Mol. Biol., Natl. Cancer Inst., Bethesda, MD, 20892, USA
JOURNAL: Gene DATE: 1989 VOLUME: 83 NUMBER: 1 PAGES: 137-46 CODEN: GENED6 ISSN: 0378-1119 LANGUAGE: English
SECTION:
CA203005 Biochemical Genetics
CA213XXX Mammalian Biochemistry
IDENTIFIERS: cDNA library cloning system insert, gene cloning system DNA insert
DESCRIPTORS:
Gene and Genetic element...
cDNA library of, construction of, cloning system for
Molecular cloning...
for cDNA library construction
Receptors...
for platelet-derived growth factor, expression of gene for, cloning system for
Gene and Genetic element,animal...
for platelet-derived growth factor receptor, expression of, cloning system for
Animal growth regulators,blood platelet-derived growth factors...
receptors for, expression of gene for, cloning system for
CAS REGISTRY NUMBERS:
125121-48-4 125121-85-9 125121-86-0 125121-87-1 125121-88-2
125121-89-3 125122-20-5 125180-30-5 as DNA insert, in construction of cDNA library, cloning system for

9/7/2 (Item 2 from file: 399)
112049430 CA: 112(7)49430k JOURNAL
PDGF induction of tyrosine phosphorylation of GTPase activating protein
AUTHOR(S): Molloy, Christopher J.; Bottaro, Donald P.; Fleming, Timothy P.; Marshall, Mark S.; Gibbs, Jackson B.; Aaronson, Stuart A.
LOCATION: Lab. Cell. Mol. Biol., Natl. Cancer Inst., Bethesda, MD, 20892, USA
JOURNAL: Nature (London) DATE: 1989 VOLUME: 342 NUMBER: 6250 PAGES: 711-14 CODEN: NATUAS ISSN: 0028-0836 LANGUAGE: English
SECTION:
CA202010 Mammalian Hormones
IDENTIFIERS: platelet growth factor GTPase activating protein, receptor kinase phosphorylation GTPase activating protein
DESCRIPTORS:
Receptors...
for platelet-derived growth factor, GTPase-activating protein phosphorylation regulation by
Cell membrane...
GTPase-activating phosphoprotein of, platelet-derived growth factor

receptor kinase in relation to
Animal growth regulators,blood platelet-derived growth factors...
GTPase-activating phosphoprotein, stimulation by, receptor mediation of
Phosphorylation,biological...
of GTPase-activating phosphoprotein, by platelet-derived growth factor receptor kinase
Proteins,specific or class, GAP (GTPase-activating protein)...
phosphorylation of, by platelet-derived growth factor receptor kinase
CAS REGISTRY NUMBERS:
60-18-4 biological studies, of GTPase activating protein, phosphorylation by platelet-derived growth factor receptor kinase
101463-26-7 GTPase-activating protein phosphorylation by

9/7/3 (Item 3 from file: 399)
112017047 CA: 112(3)17047z JOURNAL
Isolation of a novel receptor cDNA establishes the existence of two PDGF receptor genes
AUTHOR(S): Matsui, Toshimitsu; Heidaran, Mohammad; Miki, Toru; Popescu, Nicholas; La Rochelle, William; Kraus, Matthias; Pierce, Jacalyn; Aaronson, Stuart
LOCATION: Lab. Cell. Mol. Biol., Natl. Cancer Inst., Bethesda, MD, 20892, USA
JOURNAL: Science (Washington, D. C., 1883-) DATE: 1989 VOLUME: 243
NUMBER: 4892 PAGES: 800-4 CODEN: SCIEAS ISSN: 0036-8075 LANGUAGE: English
SECTION:
CA203003 Biochemical Genetics
CA202XXX Mammalian Hormones
CA213XXX Mammalian Biochemistry
IDENTIFIERS: receptor PDGF gene family human, mapping receptor PDGF gene human, cDNA platelet growth factor receptor human
DESCRIPTORS:
Chromosome,human 4...
blood platelet-derived growth factor receptor gene mapping on
Receptors...
for blood platelet-derived growth factor, of human, two isoforms of
Protein sequences...
for blood platelet-derived growth factor receptor, of human, complete
Gene and Genetic element,animal...
for blood platelet-derived growth factor receptor, of human, multiple
Animal growth regulators,blood platelet-derived growth factors...
receptors for, of human, two genes for
CAS REGISTRY NUMBERS:
124248-24-4 124248-25-5 amino acid sequence of

9/7/4 (Item 4 from file: 399)
112001387 CA: 112(1)1387z JOURNAL
Independent expression of human .alpha. or .beta. platelet-derived growth factor receptor cDNAs in a naive hematopoietic cell leads to functional coupling with mitogenic and chemotactic signaling pathways
AUTHOR(S): Matsui, Toshimitsu; Pierce, Jacalyn H.; Fleming, Timothy P.; Greenberger, Joel S.; LaRochelle, William J.; Ruggiero, Marco; Aaronson, Stuart A.
LOCATION: Lab. Cell. Mol. Biol., Natl. Cancer Inst., Bethesda, MD, 20892, USA
JOURNAL: Proc. Natl. Acad. Sci. U. S. A. DATE: 1989 VOLUME: 86
NUMBER: 21 PAGES: 8314-18 CODEN: PNASA6 ISSN: 0027-8424 LANGUAGE: English
SECTION:
CA202010 Mammalian Hormones
IDENTIFIERS: platelet growth factor receptor gene, mitogen platelet factor receptor gene chemotaxis platelet factor receptor gene blood

platelet derived growth factor
DESCRIPTORS:
Animal growth regulators,blood platelet-derived growth factors...
.alpha.- and .beta.-subunits, receptors for, expression of, chemotaxis and mitogenesis in relation to
Cell division... Chemotaxis...
blood platelet-derived growth factor subunits effect on, receptor gene expression in relation to
Receptors...
for blood platelet-derived growth factor .alpha.- and .beta.-subunits, expression of, chemotaxis and mitogenesis in relation to
Gene and Genetic element,animal...
for blood platelet-derived growth factor subunit receptors, expression of, chemotaxis and mitogenesis in relation to
Phospholipids,inositol-contg.,biological studies...
hydrolysis of, blood platelet-derived growth factor subunits effect on, receptor gene expression in relation to
CAS REGISTRY NUMBERS:
7440-70-2 properties, mobilization of, blood platelet-derived growth factor subunits effect on, receptor gene expression in relation to

9/7/5 (Item 5 from file: 399)
111225952 CA: 111(25)225952x JOURNAL
Autocrine mechanism for v-sis transformation requires cell surface localization of internally activated growth factor receptors
AUTHOR(S): Fleming, Timothy P.; Matsui, Toshimitsu; Molloy, Christopher J.; Robbins, Keith C.; Aaronson, Stuart A.
LOCATION: Lab. Cell. Mol. Biol., Natl. Cancer Inst., Bethesda, MD, 20892, USA
JOURNAL: Proc. Natl. Acad. Sci. U. S. A. DATE: 1989 VOLUME: 86
NUMBER: 20 PAGES: 8063-7 CODEN: PNASA6 ISSN: 0027-8424 LANGUAGE: English
SECTION:
CA202010 Mammalian Hormones
CA203XXX Biochemical Genetics
IDENTIFIERS: oncogene vsis growth factor receptor, blood platelet derived growth factor oncogene, transformation vsis oncogene growth factor
DESCRIPTORS:
Cell division...
blood platelet-derived growth factor-induced, receptor mediation of, oncogene v-sis transformation in relation to
Receptors...
for blood platelet-derived growth factors, of cell surface, oncogene v-sis transformation and mitogenesis in relation to
Phosphorylation,biological, auto-...
of blood platelet-derived growth factor receptors, oncogene v-sis transformation and mitogenesis in relation to
Cell membrane...
platelet-derived growth factor receptors of, oncogene v-sis transformation and mitogenesis in relation to
Animal growth regulators,blood platelet-derived growth factors...
receptors for, of cell surface, oncogene v-sis transformation and mitogenesis in relation to
Gene and Genetic element,microbial, v-sis...
transformation by, platelet-derived growth factor receptors of cell surface and mitogenesis in relation to
Transformation,neoplastic...
v-sis oncogene-induced, platelet-derived growth factor receptors of cell surface and mitogenesis in relation to

9/7/6 (Item 6 from file: 399)
111113326 CA: 111(13)113326a JOURNAL

Immunochemical localization of the epitope for a monoclonal antibody that neutralizes human platelet-derived [redacted]th factor mitogenic activity
AUTHOR(S): LaRochelle, William J.; [redacted]obbins, Keith C.; Aaronson, Stuart A.
LOCATION: Lab. Cell. Mol. Biol., Natl. Cancer Inst., Bethesda, MD, 20892, USA
JOURNAL: Mol. Cell. Biol. DATE: 1989 VOLUME: 9 NUMBER: 8 PAGES: 3538-42 CODEN: MCEBD4 ISSN: 0270-7306 LANGUAGE: English
SECTION:
CA215002 Immunochemistry
CA202XXX Mammalian Hormones
IDENTIFIERS: platelet derived growth factor mitogen epitope
DESCRIPTORS:
Animal growth regulators,blood platelet-derived growth factors...
BB homodimer, c-sis gene-encoded, epitope assocd. with mitogenic activity of human, localization of
Mitogens...
c-sis gene-encoded platelet-derived growth factor as human, epitope assocd. with, localization of
Receptors...
c-sis gene-encoded platelet-derived growth factor binding to, monoclonal antibody inhibition of, of humans, mitogenic activity in relation to
Gene and Genetic element,animal, c-sis...
platelet-derived growth factor encoded by, epitope assocd. with mitogenic activity of human, localization of

9/7/7 (Item 7 from file: 399)
111071763 CA: 111(9)71763x JOURNAL
Expression and purification of biologically active v-sis/platelet-derived growth factor B protein by using a baculovirus vector system
AUTHOR(S): Giese, Neill; May-siroff, Mary; LaRochelle, William J.; Van Wyke Coelingh, Kathleen; Aaronson, Stuart A.
LOCATION: Lab. Cel. Mol. Biol., Natl. Cancer Inst., Bethesda, MD, 20814, USA
JOURNAL: J. Virol. DATE: 1989 VOLUME: 63 NUMBER: 7 PAGES: 3080-6
CODEN: JOVIAM ISSN: 0022-538X LANGUAGE: English
SECTION:
CA202010 Mammalian Hormones
IDENTIFIERS: platelet derived growth factor subunit expression, protein vsis expression baculovirus system
DESCRIPTORS:
Virus,animal, baculo-...
as expression vector system, blood platelet-derived growth factor B protein expression in insect cells by
Animal cell line,SF9...
blood platelet-derived growth factor B protein expression in, baculovirus vector system for
Insect... Spodoptera frugiperda...
blood platelet-derived growth factor B protein expression in cells of, baculovirus vector system for
Animal growth regulators,blood platelet-derived growth factors, p28v-sis...
expression and purifn. of, in insect cell line, baculovirus vector system for
Disulfide group...
of recombinant platelet-derived growth factor B protein expressed in insect cells with baculovirus vector
Receptors...
recombinant platelet-derived growth factor B protein binding by, after expression in insect cells with baculovirus vector
Deoxyribonucleic acid formation...
recombinant platelet-derived growth factor B protein effect on

9/7/8 (Item 8 from file: 399)
103193063 CA: 103(23)193063z JOURNAL
Evidence that the v-sis gene product transforms by interaction with the receptor for platelet-derived growth factor
AUTHOR(S): Leal, Fernando; Williams, Lewis T.; Robbins, Keith C.; Aaronson, Stuart A.
LOCATION: Lab. Cell. Mol. Biol., Natl. Cancer Inst., Bethesda, MD, 20205, USA
JOURNAL: Science (Washington, D. C., 1883-) DATE: 1985 VOLUME: 230
NUMBER: 4723 PAGES: 327-30 CODEN: SCIEAS ISSN: 0036-8075 LANGUAGE: English
SECTION:
CA110005 Microbial Biochemistry
CA106XXX General Biochemistry
CA114XXX Mammalian Pathological Biochemistry
IDENTIFIERS: simian sarcoma virus gene vsis transformation, platelet growth factor receptor gene vsis
DESCRIPTORS:
Transformation,neoplastic...
by simian sarcoma virus, interaction of gene v-sis protein with receptor for platelet-derived growth factor in
Receptors...
for platelet-derived cofactor, simian sarcoma virus gene v-sis proteins interaction with, neoplastic transformation in relation to
Virus,animal, simian sarcoma...
gene v-sis protein of, interaction of, with receptors for platelet-derived growth factor, neoplastic transformation in relation to
Proteins,gene v-sis...
of simian sarcoma virus, interaction of, with receptor for platelet-derived growth factor, neoplastic transformation in relation to
Animal growth substances,blood platelet-derived growth factors...
receptor for, simian sarcoma virus gene v-sis protein interaction with, neoplastic transformation in relation to

9/7/9 (Item 1 from file: 5)
0020805795 BIOSIS Number: 89016077
AUTOCRINE MECHANISM FOR V-SIS TRANSFORMATION REQUIRES CELL SURFACE LOCALIZATION OF INTERNALLY ACTIVATED GROWTH FACTOR RECEPTORS
FLEMING T P; MATSUI T; MOLLOY C J; ROBBINS K C; AARONSON S A
LAB. CELLULAR AND MOL. BIOL., NATL. CANCER INST., NATL. INST. HEALTH, BETHESDA, MD. 20892.
PROC NATL ACAD SCI U S A 86 (20). 1989. 8063-8067. CODEN: PNASA
Language: ENGLISH
v-sis represents a prototype for the class of oncogenes that encode growth factors. Whether its platelet-derived growth factor (PDGF)-like product functionally activates its receptors within the cell or at the cell surface has potential implications in efforts to intervene with the v-sis-transformed phenotype. We demonstrate that intracellular as well as cell surface forms of two PDGF receptor gene products are tyrosine phosphorylated in v-sis transformants. In a chemically defined medium in which cell growth was dependent on v-sis expression, proliferation was partially inhibited by PDGF neutralizing antibody but completely blocked by suramin. Suramin treatment resulted in a marked reduction in tyrosine phosphorylated cell surface PDGF receptors but hod no effect on the level of tyrosine phosphorylated cell surface PDGF receptors but had no effect on the level of tyrosine phosphorylation of intracellular receptor species. All of these findings demonstrate that the v-sis-encoded mitogen can bind and activate its receptors internally but that activated receptors must achieve a cell surface location in order to functionally couple with intracellular mitogenic signaling p[redacted]ys.

9/7/10 (Item 2 from file: 5)

0020795308 BIOSIS Number: 38017526
FGF INDUCES THE PDGF-ALPHA RECEPT[illegible]ND INHIBITS MYELIN GENE EXPRESSION IN DIFFERENTIATING O-2A GLIAL PROGENITOR CELLS
MCKINNON R D; MATSUI T; AARONSON S; DUBOIS-DALCQ M
LVMP, NINDS, NIH, BETHESDA, MD. 20892.
TWENTY-NINTH ANNUAL MEETING OF THE AMERICAN SOCIETY FOR CELL BIOLOGY, HOUSTON, TEXAS, USA, NOVEMBER 5-9, 1989. J CELL BIOL 109 (4 PART 2). 1989. 97A. - CODEN: JCLBA
Language: ENGLISH

9/7/11 (Item 3 from file: 5)
0020794674 BIOSIS Number: 38016892
OVEREXPRESSION OF PROTO-ONCOGENES ENCODING GROWTH FACTOR OR GROWTH FACTOR RECEPTOR PROTEINS IN MALIGNANCY
KRAUS M H; DI FIORE P P; PIERCE J H; ROBBINS K C; AARONSON S A
NATL. CANCER INST., BETHESDA, MD. 20892, USA.
FURTH, M. AND M. GREAVES (ED.). CANCER CELLS COLD SPRING HARBOR, VOL. 7. MOLECULAR DIAGNOSTICS OF HUMAN CANCER; CONFERENCE, COLD SPRING HARBOR, NEW YORK, USA. XXII+414P. COLD SPRING HARBOR LABORATORY PRESS: COLD SPRING HARBOR, NEW YORK, USA. ILLUS. PAPER. ISBN 0-87969-324-X. 0 (0). 1989. 303-310. CODEN: CACEE
Language: ENGLISH

9/7/12 (Item 4 from file: 5)
0020172789 BIOSIS Number: 88084979
IMMUNOCHEMICAL LOCALIZATION OF THE EPITOPE FOR A MONOCLONAL ANTIBODY THAT NEUTRALIZES HUMAN PLATELET-DERIVED GROWTH FACTOR MITOGENIC ACTIVITY
LAROCHELLE W J; ROBBINS K C; AARONSON S A
LAB. CELL. MOL. BIOL., NATL. CANCER INST., BETHESDA, MD. 20892, USA.
MOL CELL BIOL 9 (8). 1989. 3538-3542. CODEN: MCEBD
Language: ENGLISH
A monoclonal antibody (mAb), sis 1, generated against human c-sis-encoded platelet-derived growth factor (PDGF) BB, was shown by enzyme-linked immunosorbent assay and Western blot (immunoblot) analysis to recognize human PDGF BB and human platelet PDGF AB but not the human PDGF AA. This monoclonal antibody potently inhibited PDGF receptor-binding and mitogenic activities of both human PDGF BB and PDGF AB but had no effect on PDGF AA. Finally, we demonstrated that an immunoaffinity-purified anti-c-sis peptide antibody (anti-V4) which also blocked binding of PDGF BB to its cognate receptor and competed with mAb sis 1 for binding to PDGF BB. All of these results suggest that mAb sis 1 recognizes an epitope of the c-sis gene product, PDGF BB, that spatially overlaps the V4 surface domain of PDGF BB, immunochemically localizing a region of PDGF BB critical for PDGF receptor binding and activation.

9/7/13 (Item 5 from file: 5)
0019593319 BIOSIS Number: 88049351
EXPRESSION AND PURIFICATION OF BIOLOGICALLY ACTIVE V-SIS PLATELET-DERIVED GROWTH FACTOR B PROTEIN BY USING A BACULOVIRUS VECTOR SYSTEM
GIESE N; MAY-SIROFF M; LAROCHELLE W J; VAN WYKE COELINGH K; AARONSON S A
LAB. CELLULAR AND MOLECULAR BIOL., BUILDING 37, ROOM 1E24, NATL. CANCER INST., BETHESDA, MD. 20814.
J VIROL 63 (7). 1989. 3080-3086. CODEN: JOVIA
Language: ENGLISH
Malignant transformation induced by simian sarcoma virus is mediated by its v-sis protein, the monkey homolog of the platelet-derived growth factor (PDGF) B chain. By use of an appropriately engineered baculovirus expression vector, the v-sis protein was expressed in the insect cell line Spodoptera frugiperda (Sf9) at a level 50- to 100-fold higher than that observed with overexpression in mammalian-cell transfectants. The sis protein produced by Sf9 cells underwent processing similar to that observed in mammalian cells, including efficient disulfide-linked dimer formation. Moreover, the recombinant sis protein was capable of binding PDGF receptors and inducing DNA synthesis as efficiently as PDGF-B synthesized by mammalian cells. A significant fraction of sis protein was released from Sf9 cells which made possible a one-step immunoaffinity purification to

near homogeneity with a 40% recovery of biological activity. These results demonstrate that a protein whose normal processing requires both intrachain and interchain disulfide-bridge formation can be efficiently expressed in a biologically active form in insect cells by using a baculovirus vector system.

9/7/14 (Item 6 from file: 5)
0019191915 BIOSIS Number: 87092819
ISOLATION OF A NOVEL RECEPTOR COMPLEMENTARY DNA ESTABLISHES THE EXISTENCE OF TWO PDGF RECEPTOR GENES
MATSUI T; HEIDARAN M; MIKI T; POPESCU N; LA ROCHELLE W; KRAUS M; PIERCE J ; AARONSON S
LAB. CELL. MOL. BIOL., NATL. CANCER INST., BETHESDA, MD. 20892.
SCIENCE (WASHINGTON D C) 243 (4892). 1989. 800-804. CODEN: SCIEA
Language: ENGLISH
A genomic sequence and cloned complementary DNA has been identified for a novel receptor-like gene of the PDGF receptor/CSF1 receptor subfamily (platelet-derived growth factor receptor/colony-stimulating factor type 1 receptor). The gene recognized a 6.4-kilobase transcript that was coexpressed in normal human tissues with the 5.3-kilobase PDGF receptor messenger RNA. Introduction of complementary DNA of the novel gene into COS-1 cells led to expression of proteins that were specifically detected with antiserum directed against a predicted peptide. When the new gene was transfected into COS-1 cells, a characteristic pattern of binding of the PDGF isoforms was observed, which was different from the pattern observed with the known PDGF receptor. Tyrosine phosphorylation of the receptor in response to the PDGF isoforms was also different from the known receptor. The new PDGF receptor gene was localized to chromosome 4q11-4q12. The existence of genes encoding two PDGF receptors that interact in a distinct manner with three different PDGF isoforms likely confers considerable regulatory flexibility in the functional responses to PDGF.
?t s9/7/15-35

9/7/15 (Item 7 from file: 5)
0019170827 BIOSIS Number: 87082093
FUNCTIONAL IDENTIFICATION OF REGULATORY ELEMENTS WITHIN THE PROMOTER REGION OF PLATELET-DERIVED GROWTH FACTOR 2
PECH M; RAO C D; ROBBINS K C; AARONSON S A
LAB. OF CELL. AND MOL. BIOL., NATL. CANCER INST., BETHESDA, MD. 20892.
MOL CELL BIOL 9 (2). 1989. 396-405. CODEN: MCEBD
Language: ENGLISH
Human platelet-derived growth factor (PDGF) is composed of two polypeptide chains, PDGF-1 and PDGF-2, the human homolog of the v-sis oncogene. Deregulation of PDGF-2 expression can confer a growth advantage to cells possessing the cognate receptor and, thus, may contribute to the malignant phenotype. We investigated the regulation of PDGF-2 mRNA expression during megakaryocytic differentiation of K562 cells. Induction by 12-O-tetradecanoylphorbol-13-acetate (TPA) led to a greater than 200-fold increase in PDGF-2 transcript levels in these cells. Induction was dependent on protein synthesis and was not enhanced by cycloheximide exposure. In our initial investigation of the PDGF-2 promoter, a minimal promoter region, which included sequences extending only 42 base pairs upstream of the TATA signal, was found to be as efficient as 4 kilobase pairs upstream of the TATA signal in driving expression of a reporter gene in uninduced K562 cells. We also functionally identified different regulatory sequence elements of the PDGF-2 promoter in TPA-induced K562 cells. One region acted as a transcriptional silencer, while another region was necessary for maximal activity of the promoter in megakaryoblasts. This region was shown to bind nuclear factors and was the target for trans-activation in normal and tumor cells. In one tumor line, which expressed high PDGF-2 mRNA levels, the presence of the positive regulatory region resulted in a 30-fold increase in promoter activity. However, the ability of the minimal PDGF-2 promoter to drive reporter gene expression in uninduced K562 cells and normal fi[redacted]asts, which contained no detectable PDGF-2 transcripts, implies the [redacted]stence of other negative control mechanisms beyond the regulation of promoter activity.

9/7/16 (Item 8 from file: 5)
0017622465 BIOSIS Number: 84055970
THE HUMAN TRANSFORMING GROWTH FACTOR TYPE ALPHA CODING SEQUENCE IS NOT A DIRECT-ACTING ONCOGENE WHEN OVEREXPRESSED IN NIH 3T3 CELLS
FINZI E; FLEMING T; SEGATTO O; PENNINGTON C Y; BRINGMAN T S; DERYNCK R; AARONSON S A
LAB. CELL. MOL. BIOL., NATL. CANCER INST., BUILD. 37, ROOM 1E24, BETHESDA, MD. 20892.
PROC NATL ACAD SCI U S A 84 (11). 1987. 3733-3737. CODEN: PNASA
Language: ENGLISH
A papetide secreted by some tumor cells in vitro imparts anchorage-independent growth to normal rat kidney (NRK) cells and has been termed transforming growth factor type .alpha. (TGF-.alpha.). To directly investigate the transforming properties of this factor, the human sequence coding for TGF-.alpha. was placed under the control of either a metallothionein promoter or a retroviral long terminal repeat. These constructs failed to induce morphological transformation upon transfection of NIH 3T3 cells, whereas viral oncogenes encoding a truncated form of its cognate receptor, the EGF receptor, or another growth factor, sis/platelet-derived growth factor 2, efficiently induced transformed foci. When NIH 3T3 clonal sublines were selected by transfection of TGF-.alpha. expression vectors in the presence of a dominant selectable marker, they were shown to secrete large amounts of TGF-.alpha. into the medium, to have downregulated EGF receptors, and to be inhibited in growth by TGF-.alpha. monoclonal antibody. These results indicated that secreted TGF-.alpha. interacts with its receptor at a cell surface location. Single cell-derived TGF-.alpha.-expressing sublines grew to high saturation density in culture. However, when plated as single cells on contact-inhibited monolayers of NIH 3T3 cells, they failed to form colonies, whereas v-sis- and v-erbB-transfected cells formed transformed colonies under the same conditions. Moreover, TGF-.alpha.-expressing sublines were not tumorigenic in nude mice. These and other results imply that TGF-.alpha. exerts a growth-promoting effect on the entire NIH 3T3 cell population after secretion into the medium but little, if any, effect on the individual cell synthesizing this factor. It is concluded that the normal coding sequence for TGF-.alpha. is not a direct-acting oncogene when overexpressed in NIH 3T3 cells.

9/7/17 (Item 9 from file: 5)
0017603432 BIOSIS Number: 84047863
THE ROLE OF INDIVIDUAL CYSTEINE RESIDUES IN THE STRUCTURE AND FUNCTION OF THE V-SIS GENE PRODUCT
GIESE N A; ROBBINS K C; AARONSON S A
LAB. CELLULAR AND MOLECULAR BIOL., NATL. CANCER INST., NATL. INST. HEALTH, BETHESDA, MD. 20892.
SCIENCE (WASH D C) 236 (4806). 1987. 1315-1318. CODEN: SCIEA
Language: ENGLISH
The v-sis oncogene encodes a platelet-derived growth factor (PDGF)-related product whose transforming activity is mediated by its functional interaction with the PDGF receptor. PDGF, as well as processed forms of the v-sis gene product, is a disulfide-linked dimer with eight conserved cysteine residues in the minimum region necessary for biologic activity. Site-directed mutagenesis of the v-sis gene revealed that each conserved cysteine residue was required directly or indirectly for disulfide-linked dimer formation. However, substitution of serine for cysteine codons at any of four positions had no detrimental effect on transforming activity of the encoded v-sis protein. These results establish that interchain disulfide bonds are not essential in order for this protein to act as a functional ligand for the PDGF receptor. The remaining four substitutions of serine for cysteine each inactivated transforming function of the molecule. In each case this was associated with loss of a conformation shown to involve intramolecular disulfide bonds. These studies provide insight into the role of individual cysteine residues in determining the structure of the sis/PDGF molecule critical for biological activity.

9/7/18 (Item 10 from file: 5)
0017049888 BIOSIS Number: 32029976
ONCOGENES WHICH CODE FOR GROWTH FACTORS OR THEIR RECEPTORS
AARONSON S A
NATL. CANCER INST., BETHESDA, MD, USA.
UICC (UNION INTERNATIONALE CONTRE LE CANCER, INTERNATIONAL UNION AGAINST CANCER). 14TH INTERNATIONAL CANCER CONGRESS, BUDAPEST, HUNGARY, AUG. 21-27, 1986. ABSTRACTS, LECTURES, SYMPOSIA AND FREE COMMUNICATIONS, VOLS. 1, 2, 3, LATE ABSTRACTS, AND REGISTER. XVI+479P.(VOL. 1); XVI+298P.(VOL. 2); XVI+531P.(VOL. 3); 15P.(LATE ABSTRACTS); 40P.(REGISTER) S. KARGER AG: BASEL, SWITZERLAND; NEW YORK, N.Y., USA; AKADEMIAI KIADO: BUDAPEST, HUNGARY. PAPER. ISBN 3-8055-4434-0(KARGER); ISBN 963-05-4422-9(VOL. 1); ISBN 963-05-4423-7(VOL. 2); ISBN 963-05-4424-5(VOL. 3); ISBN 963-05-4439-3(LATE ABSTRACTS); ISBN 963-05-4425-3(REGISTER); ISBN 963-05-4421-0(GENERAL). 0 (0). 1986. 237. CODEN: 24757
Language: ENGLISH

9/7/19 (Item 11 from file: 5)
0016056068 BIOSIS Number: 81025006
EVIDENCE THAT THE V-SIS GENE PRODUCT TRANSFORMS BY INTERACTION WITH THE RECEPTOR FOR PLATELET-DERIVED GROWTH FACTOR
LEAL F; WILLIAMS L T; ROBBINS K C; AARONSON S A
LAB. CELLULAR MOLECULAR BIOL., NATL. CANCER INST., BETHESDA, MD. 20205.
SCIENCE (WASH D C) 230 (4723). 1985. 327-330. CODEN: SCIEA
Language: ENGLISH
A scheme for partial purification of biological active v-sis-coded protein from cells transformed with simian sarcoma virus (SSV) has made possible a functional comparison of the transforming protein with platelet-derived growth factor (PDGF). The SSV-transforming gene product is capable of specifically bindng PDGF receptors, stimulating tyrosine phosphorylation of PDGF receptors, and inducing DNA synthesis in quiescent fibroblasts. Each of these activities was specifically inhibited by antibodies to different regions of the v-sis gene product. Moreover, viral infection of a variety of cell types revealed a strict correlation between those cells possessing PDGF receptors and those susceptible to transformation by SSV. These findings provide evidence that SSV-transforming activity is mediated by the interaction of a virus-coded mitogen with PDGF receptors.

9/7/20 (Item 1 from file: 155)
07174848 90081848
PDGF induction of tyrosine phosphorylation of GTPase activating protein.
Molloy CJ; Bottaro DP; Fleming TP; Marshall MS; Gibbs JB; Aaronson SA
Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, Maryland 20892.
Nature (ENGLAND) Dec 7 1989, 342 (6250) p711-4, ISSN 0028-0836
Journal Code: NSC
Languages: ENGLISH
The cascade of biochemical events triggered by growth factors and their receptors is central to understanding normal cell-growth regulation and its subversion in cancer. Ras proteins (p21ras) have been implicated in signal transduction pathways used by several growth factors, including platelet-derived growth factor (PDGF). These guanine nucleotide-binding Ras proteins specifically interact with a cellular GTPase-activating protein (GAP). Here we report that in intact quiescent fibroblasts, both AA and BB homodimers of PDGF rapidly induce tyrosine phosphorylation of GAP under conditions in which insulin and basic fibroblast growth factor (bFGF) are ineffective. Although GAP is located predominantly in the cytosol, most tyrosine-phosphorylated GAP is associated with the cell membrane, the site of p21ras biological activity. These results provide a direct biochemical link between activated PDGF-receptor tyrosine kinases and the p21ras-GAP mitogenic signalling system.

9/7/21 (Item 2 from file: 155)
07139797 90044797

Independent expression of human alpha or beta platelet-derived growth factor receptor cDNAs in a naiv[redacted]matopoietic cell leads to functional coupling with mitogenic and chemotactic signaling pathways.

Matsui T; Pierce JH; Fleming TP; Greenberger JS; LaRochelle WJ; Ruggiero M; Aaronson SA

Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, MD 20892.

Proc Natl Acad Sci U S A (UNITED STATES) Nov 1989, 86 (21) p8314-8, ISSN 0027-8424 Journal Code: PV3

Languages: ENGLISH

Distinct genes encode alpha and beta platelet-derived growth factor (PDGF) receptors that differ in their abilities to be triggered by three dimeric forms of the PDGF molecule. We show that PDGF-receptor mitogenic function can be reconstituted in a naive hematopoietic cell line by introduction of expression vectors for either alpha or beta PDGF receptor cDNAs. Thus, each receptor is independently capable of coupling with mitogenic signal-transduction pathways inherently present in these cells. Activation of either receptor also resulted in chemotaxis, alterations in inositol lipid metabolism, and mobilization of intracellular Ca2+. The magnitude of these functional responses correlated well with the binding properties of the different PDGF isoforms to each receptor. Thus, availability of specific PDGF isoforms and relative expression of each PDGF-receptor gene product are major determinants of the spectrum of known PDGF responses.

9/7/22 (Item 3 from file: 155)
07139738 90046738

Autocrine mechanism for v-sis transformation requires cell surface localization of internally activated growth factor receptors.

Fleming TP; Matsui T; Molloy CJ; Robbins KC; Aaronson SA

Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, MD 20892.

Proc Natl Acad Sci U S A (UNITED STATES) Oct 1989, 86 (20) p8063-7, ISSN 0027-8424 Journal Code: PV3

Languages: ENGLISH

v-sis represents a prototype for the class of oncogenes that encode growth factors. Whether its platelet-derived growth factor (PDGF)-like product functionally activates its receptors within the cell or at the cell surface has potential implications in efforts to intervene with the v-sis-transformed phenotype. We demonstrate that intracellular as well as cell surface forms of two PDGF receptor gene products are tyrosine phosphorylated in v-sis transformants. In a chemically defined medium in which cell growth was dependent on v-sis expression, proliferation was partially inhibited by PDGF neutralizing antibody but completely blocked by suramin. Suramin treatment resulted in a marked reduction in tyrosine phosphorylated cell surface PDGF receptors but had no effect on the level of tyrosine phosphorylation of intracellular receptor species. All of these findings demonstrate that the v-sis-encoded mitogen can bind and activate its receptors internally but that activated receptors must achieve a cell surface location in order to functionally couple with intracellular mitogenic signaling pathways.

9/7/23 (Item 4 from file: 155)
07107811 90014811

Immunochemical localization of the epitope for a monoclonal antibody that neutralizes human platelet-derived growth factor mitogenic activity.

LaRochelle WJ; Robbins KC; Aaronson SA

Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, Maryland 20892.

Mol Cell Biol (UNITED STATES) Aug 1989, 9 (8) p3538-42, ISSN 0270-7306 Journal Code: NGY

Languages: ENGLISH

A monoclonal antibody (mAb), sis 1, generated against human c-sis-encoded platelet-derived growth factor (PDGF) BB, was shown by enzyme-linked immunosorbent assay and Western blot (immunoblot) analysis to recognize human PDGF BB and human platelet PDGF AB but not the human PDGF AA. This

monoclonal antibody potently inhibited PDGF receptor-binding and mitogenic activities of both human PDGF BB and PDGF AB but had no effect on PDGF AA. Finally, we demonstrated that an immunoaffinity-purified anti-c-sis peptide antibody (anti-V4) which also blocked binding of PDGF BB to its cognate receptor and competed with mAb sis 1 for binding to PDGF BB. All of these results suggest that mAb sis 1 recognizes an epitope of the c-sis gene product, PDGF BB, that spatially overlaps the V4 surface domain of PDGF BB, immunochemically localizing a region of PDGF BB critical for PDGF receptor binding and activation.

9/7/24 (Item 5 from file: 155)
06957102 89259102

Expression and purification of biologically active v-sis/platelet-derived growth factor B protein by using a baculovirus vector system.

Giese N; May-Siroff M; LaRochelle WJ; van Wyke Coelingh K; Aaronson SA

Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, Maryland 20814.

J Virol Jul 1989, 63 (7) p3080-6, ISSN 0022-538X Journal Code: KCV

Languages: ENGLISH

Malignant transformation induced by simian sarcoma virus is mediated by its v-sis protein, the monkey homolog of the platelet-derived growth factor (PDGF) B chain. By use of an appropriately engineered baculovirus expression vector, the v-sis protein was expressed in the insect cell line Spodoptera frugiperda (Sf9) at a level 50- to 100-fold higher than that observed with overexpression in mammalian-cell transfectants. The sis protein produced by Sf9 cells underwent processing similar to that observed in mammalian cells, including efficient disulfide-linked dimer formation. Moreover, the recombinant sis protein was capable of binding PDGF receptors and inducing DNA synthesis as efficiently as PDGF-B synthesized by mammalian cells. A significant fraction of sis protein was released from Sf9 cells, which made possible a one-step immunoaffinity purification to near homogeneity with a 40% recovery of biological activity. These results demonstrate that a protein whose normal processing requires both intrachain and interchain disulfide-bridge formation can be efficiently expressed in a biologically active form in insect cells by using a baculovirus vector system.

9/7/25 (Item 6 from file: 155)
06924834 89226834

Mechanisms by which genes encoding growth factors and growth factor receptors contribute to malignant transformation.

Kraus MH; Pierce JH; Fleming TP; Robbins KC; Di Fiore PP; Aaronson SA

National Cancer Institute, Laboratory of Cellular and Molecular Biology, Bethesda, Maryland 20892.

Ann N Y Acad Sci 1988, 551 p320-35; discussion 336, ISSN 0077-8923 Journal Code: 5NM

Languages: ENGLISH

Document Type: Review

(52 Refs.)

9/7/26 (Item 7 from file: 155)
06917031 89219031

Functional identification of regulatory elements within the promoter region of platelet-derived growth factor 2.

Pech M; Rao CD; Robbins KC; Aaronson SA

Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, Maryland 20892.

Mol Cell Biol Feb 1989, 9 (2) p396-405, ISSN 0270-7306 Journal Code: NGY

Languages: ENGLISH

Human platelet-derived growth factor (PDGF) is composed of two polypeptide chains, PDGF-1 and PDGF-2, the human homolog of the v-sis oncogene. Deregulation of PDGF-2 expression can confer a growth advantage to cells possessing the cognate r[illegible]or and, thus, may contribute to the malignant phenotype. We investigated the regulation of PDGF-2 mRNA expression during megakaryocytic differentiation of K562 cells. Induction

by 12-O-tetradecanoylphorbol-13-acetate (TPA) led to a greater than 200-fold increase in PDGF-2 transcript levels in these cells. Induction was dependent on protein synthesis and was not enhanced by cycloheximide exposure. In our initial investigation of the PDGF-2 promoter, a minimal promoter region, which included sequences extending only 42 base pairs upstream of the TATA signal, was found to be as efficient as 4 kilobase pairs upstream of the TATA signal in driving expression of a reporter gene in uninduced K562 cells. We also functionally identified different regulatory sequence elements of the PDGF-2 promoter in TPA-induced K562 cells. One region acted as a transcriptional silencer, while another region was necessary for maximal activity of the promoter in megakaryoblasts. This region was shown to bind nuclear factors and was the target for trans-activation in normal and tumor cells. In one tumor cell line, which expressed high PDGF-2 mRNA levels, the presence of the positive regulatory region resulted in a 30-fold increase in promoter activity. However, the ability of the minimal PDGF-2 promoter to drive reporter gene expression in uninduced K562 cells and normal fibroblasts, which contained no detectable PDGF-2 transcripts, implies the existence of other negative control mechanisms beyond the regulation of promoter activity.

9/7/27 (Item 8 from file: 155)
06828149 89130149

Isolation of a novel receptor cDNA establishes the existence of two PDGF receptor genes.

Matsui T; Heidaran M; Miki T; Popescu N; La Rochelle W; Kraus M; Pierce J ; Aaronson S

Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, MD 20892.

Science Feb 10 1989, 243 (4892) p800-4, ISSN 0036-8075 Journal Code: UJ7

Languages: ENGLISH

A genomic sequence and cloned complementary DNA has been identified for a novel receptor-like gene of the PDGF receptor/CSF1 receptor subfamily (platelet-derived growth factor receptor/colony-stimulating factor type 1 receptor). The gene recognized a 6.4-kilobase transcript that was coexpressed in normal human tissues with the 5.3-kilobase PDGF receptor messenger RNA. Introduction of complementary DNA of the novel gene into COS-1 cells led to expression of proteins that were specifically detected with antiserum directed against a predicted peptide. When the new gene was transfected into COS-1 cells, a characteristic pattern of binding of the PDGF isoforms was observed, which was different from the pattern observed with the known PDGF receptor. Tyrosine phosphorylation of the receptor in response to the PDGF isoforms was also different from the known receptor. The new PDGF receptor gene was localized to chromosome 4q11-4q12. The existence of genes encoding two PDGF receptors that interact in a distinct manner with three different PDGF isoforms likely confers considerable regulatory flexibility in the functional responses to PDGF.

9/7/28 (Item 9 from file: 155)
06819973 89121973

Involvement of oncogene-coded growth factors in the neoplastic process.

Robbins KC; King CR; Giese NA; Leal F; Igarashi H; Aaronson SA

Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, Maryland 20892.

Gene Amplif Anal 1986, 4 p161-76, ISSN 0275-2778 Journal Code: GEF

Languages: ENGLISH

Document Type: Review

(44 Refs.)

9/7/29 (Item 10 from file: 155)
06685726 88330726

Role of genes for normal growth factors in human malignancy.

Aaronson SA; Igarashi H; Rao CD; Finzi E; Fleming TP; Segatto O; Robbins KC

Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, Maryland 20892

Int Symp Princess Takamatsu Cancer Res Fund 1986, 17 p95-108,
Journal Code: HHI
Languages: ENGLISH
Document Type: Review

The human homologue of the v-sis oncogene encodes one chain of human platelet-derived growth factor (PDGF-2). Previous studies have shown that expression of the coding sequence for this growth factor induces malignant transformation of NIH/3T3 cells. We demonstrate the detection of sis/PDGF-2 products indistinguishable from functional PDGF-2 homodimers in human tumor cells. These findings support the concept that expression of the sis/PDGF-2 product in human cells responsive to its proliferative actions can be an important step in the processes leading to malignancy. Unlike sis/PDGF-2, which remains tightly cell-associated, another growth factor, termed transforming growth factor alpha (TGF alpha), is actively secreted. Expression vectors for the TGF alpha coding sequence failed to induce primary transformed foci upon transfection of NIH/3T3 cells despite high levels of TGF alpha synthesized by these cells. Moreover, transfectants selected for secretion of high levels of TGF alpha failed to form colonies on contact inhibited NIH/3T3 monolayers or to induce tumors upon inoculation of nude mice. Thus, the ability of a growth factor to induce autonomous in vitro or in vivo growth is dependent upon more than expression of cognate receptors by the cell in which it is synthesized. (46 Refs.)

9/7/30 (Item 11 from file: 155)
06676698 88321698

Comparison of biological properties and transforming potential of human PDGF-A and PDGF-B chains.

Beckmann MP; Betsholtz C; Heldin CH; Westermark B; Di Marco E; Di Fiore PP; Robbins KC; Aaronson SA

Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, MD 20892.

Science Sep 9 1988, 241 (4871) p1346-9, ISSN 0036-8075
Journal Code: UJ7
Languages: ENGLISH

Human platelet-derived growth factor (PDGF) consists of two distinct but related polypeptide chains designated PDGF-A and PDGF-B. The gene encoding PDGF-B has given rise to the v-sis oncogene. In the present study the transforming activities of PDGF-A and PDGF-B genes are compared. The PDGF-A chain gene is markedly less efficient in inducing transformation than the PDGF-B gene under the influence of the same promoter. There are significant differences in the secretory and growth stimulating properties of the two chains. These properties appear to account for the much more potent transforming ability of the PDGF-B gene. These findings provide insights into biologic properties of a growth factor responsible for potent autocrine stimulation of abnormal cell proliferation.

9/7/31 (Item 12 from file: 155)
06260338 87234338

The role of individual cysteine residues in the structure and function of the v-sis gene product.

Giese NA; Robbins KC; Aaronson SA

Science Jun 5 1987, 236 (4806) p1315-8, ISSN 0036-8075
Journal Code: UJ7
Languages: ENGLISH

The v-sis oncogene encodes a platelet-derived growth factor (PDGF)-related product whose transforming activity is mediated by its functional interaction with the PDGF receptor. PDGF, as well as processed forms of the v-sis gene product, is a disulfide-linked dimer with eight conserved cysteine residues in the minimum region necessary for biologic activity. Site-directed mutagenesis of the v-sis gene revealed that each conserved cysteine residue was required directly or indirectly for disulfide-linked dimer formation. However, substitution of serine for cysteine codons at any of four positions had no detrimental effect on transforming activity of the encoded v-sis protein. These results establish that intra-chain disulfide bonds are not essential in order for this protein

to act as a functional ligand for the PDGF receptor. The remaining four substitutions of serine for cysteine each inactivated transforming function of the molecule. In each case this was associated with loss of a conformation shown to involve intramolecular disulfide bonds. These studies provide insight into the role of individual cysteine residues in determining the structure of the sis/PDGF molecule critical for biological activity.

9/7/32 (Item 13 from file: 155)
06257891 87231891

The human transforming growth factor type alpha coding sequence is not a direct-acting oncogene when overexpressed in NIH 3T3 cells.

Finzi E; Fleming T; Segatto O; Pennington CY; Bringman TS; Derynck R; Aaronson SA

Proc Natl Acad Sci U S A Jun 1987, 84 (11) p3733-7, ISSN 0027-8424
Journal Code: PV3
Languages: ENGLISH

A peptide secreted by some tumor cells in vitro imparts anchorage-independent growth to normal rat kidney (NRK) cells and has been termed transforming growth factor type alpha (TGF-alpha). To directly investigate the transforming properties of this factor, the human sequence coding for TGF-alpha was placed under the control of either a metallothionein promoter or a retroviral long terminal repeat. These constructs failed to induce morphological transformation upon transfection of NIH 3T3 cells, whereas viral oncogenes encoding a truncated form of its cognate receptor, the EGF receptor, or another growth factor, sis/platelet-derived growth factor 2, efficiently induced transformed foci. When NIH 3T3 clonal sublines were selected by transfection of TGF-alpha expression vectors in the presence of a dominant selectable marker, they were shown to secrete large amounts of TGF-alpha into the medium, to have downregulated EGF receptors, and to be inhibited in growth by TGF-alpha monoclonal antibody. These results indicated that secreted TGF-alpha interacts with its receptor at a cell surface location. Single cell-derived TGF-alpha-expressing sublines grew to high saturation density in culture. However, when plated as single cells on contact-inhibited monolayers of NIH 3T3 cells, they failed to form colonies, whereas v-sis- and v-erbB-transfected cells formed transformed colonies under the same conditions. Moreover, TGF-alpha-expressing sublines were not tumorigenic in nude mice. These and other results imply that TGF-alpha exerts a growth-promoting effect on the entire NIH 3T3 cell population after secretion into the medium but little, if any, effect on the individual cell synthesizing this factor. It is concluded that the normal coding sequence for TGF-alpha is not a direct-acting oncogene when overexpressed in NIH 3T3 cells.

9/7/33 (Item 14 from file: 155)
05792655 86093655

Human tumor cell lines with EGF receptor gene amplification in the absence of aberrant sized mRNAs.

King CR; Kraus MH; Williams LT; Merlino GT; Pastan IH; Aaronson SA
Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, MD 20892.

Nucleic Acids Res Dec 9 1985, 13 (23) p8477-86, ISSN 0305-1048
Journal Code: 08L
Languages: ENGLISH

A survey of human tumor cell lines for increased PDGF or EGF receptors identified 5 lines which bind from 6 to 13 times more EGF than normal human fibroblasts. Immunoprecipitation analysis links the elevated binding activity to increased quantities of the EGF receptor protein. EGF receptor gene amplification was detected in 2 of the cell lines. No evidence for EGF receptor gene rearrangements was found at the level of DNA or RNA structure. The results suggest that elevated levels of EGF receptor can be associated with at least three distinct mechanisms. These include gene amplification accompanied by rearrangement, gene amplification without accompanied alteration of mRNA transcripts, and extensive expression without gene amplification

9/7/34 (Item 15 from file: 155)
05752292 86053292
Transforming genes of human malignancies.
Aaronson SA; Tronick SR
Laboratory of Cellular and Molecular Biology, National Cancer Institute, Bethesda, Maryland 20205.
Carcinog Compr Surv 1985, 10 p35-49, ISSN 0147-4006 Journal Code: CNU
Languages: ENGLISH
Document Type: Review
(74 Refs.)

9/7/35 (Item 16 from file: 155)
05717835 86018835
Evidence that the v-sis gene product transforms by interaction with the receptor for platelet-derived growth factor.
Leal F; Williams LT; Robbins KC; Aaronson SA
Science Oct 18 1985, 230 (4723) p327-30, ISSN 0036-8075 Journal Code: UJ7
Languages: ENGLISH
A scheme for partial purification of biologically active v-sis-coded protein from cells transformed with simian sarcoma virus (SSV) has made possible a functional comparison of the transforming protein with platelet-derived growth factor (PDGF). The SSV-transforming gene product is capable of specifically binding PDGF receptors, stimulating tyrosine phosphorylation of PDGF receptors, and inducing DNA synthesis in quiescent fibroblasts. Each of these activities was specifically inhibited by antibodies to different regions of the v-sis gene product. Moreover, viral infection of a variety of cell types revealed a strict correlation between those cells possessing PDGF receptors and those susceptible to transformation by SSV. These findings provide evidence that SSV-transforming activity is mediated by the interaction of a virus-coded mitogen with PDGF receptors.

?t s19/7/1-27

19/7/1 (Item 1 from file: 5)
0019024207 BIOSIS Number: 87012207
KIDNEY EPITHELIAL CELLS EXPRESS C-SIS PROTOONCOGENE AND SECRET PDGF-LIKE PROTEIN
KARTHA S; BRADHAM D M; GROTENDORST G R; TOBACK F G
DEP. MED., BOX 453, UNIV. CHICAGO, 5841 SOUTH MARYLAND AVE., CHICAGO, ILL. 60637.
AM J PHYSIOL 255 (4 PART 2). 1988. F800-F806. CODEN: AJPHA
Language: ENGLISH
Nontransformed monkey kidney cells (BSC-1 line), used as a model for renal epithelium, were assayed for release of platelet-derived growth factor (PDGF)-like proteins. BSC-1 cells continuously released a mitogenic activity for fibroblasts and a chemoattractant activity for smooth muscle cells, each of which was inhibited 90-90% by an antibody to human PDGF. A

cDNA probe for the PDGF B-chain gene (c-sis), but not for the A-chain gene, hybridized to mRNA obtained from growing and quiescent cells. c-sis gene expression and PDGF-like protein secretion were studied in the presence of known growth-regulatory molecules. A secreted BSC-1 cell protein identical to transforming growth factor .beta.2 inhibited DNA synthesis in growing cultures and induced marked accumulation of c-sis mRNA without a corresponding increase in the release of PDGF-like activity. Adenosine diphosphate stimulated DNA synthesis in quiescent cultures and enhanced both c-cis expression and release of PDGF-like activity. However, growing the quiescent cells did not express the PDGF receptor gene or exhibit a mitogenic response to authentic PDGF. Thus the PDGF-like protein released by these kidney epithelial cells could contribute to growth control by a paracrine mechanism.

19/7/2 (Item 2 from file: 5)
0018754572 BIOSIS Number: 86126358
ROLE OF IIB-IIIA-LIKE GLYCOPROTEINS IN CELL-SUBSTRATUM ADHESION OF HUMAN MELANOMA CELLS
KNUDSEN K A; SMITH L; SMITH S; KARCZEWSKI J; TUSZYNSKI G P
LANKENAU MED. RES. CENT., PHILADELPHIA, PA. 19151.
J CELL PHYSIOL 136 (3). 1988. 471-478. CODEN: JCLLA
Language: ENGLISH
The platelet fibrinogen receptor, glycoprotein complex IIb-IIIa, was isolated from human platelets by lectin and monoclonal antibody affinity chromatography and a polyclonal antiserum (anti-IIb-IIIa) was generated and used to probe for the presence and function of IIb-IIIa-like molecules in two adherent human cell lines. Both C32 melanoma cells and W138 fibroblasts expressed a IIb-IIIa-like complex on their surface as indicated by immunoprecipitation of detergent extracts of surface radiolabeled cells. When added to cells plated in medium containing 10% serum, the anti-IIb-IIIa antiserum perturbed the adhesion of C32 melanoma cells, but not of W138 fibroblasts. In a serum-free system, anti-IIb-IIIa antibodies inhibited attachment and spreading of C32 cells to fibrinogen, vitronectin, and fibronectin adsorbed to glass. Anti-IIb-IIIa had no effect on the attachment and spreading of W138 cells to the extracellular matrix proteins, however. Thus, the IIb-IIIa-like complex appears to play a predominant role in cell-substratum adhesion of C32 cells, but not W138 cells, and may result from the fact that, on a protein basis, the C32 melanoma cells express approximately 3 times more complex on their surface than do W138 fibroblasts. The results suggest that the relative abundance of a particular adhesion receptor on the cell surface may govern its importance to cell-substratum adhesion.

19/7/3 (Item 3 from file: 5)
0018537822 BIOSIS Number: 86015616
MOLECULAR CLONING OF THE ALPHA SUBUNIT OF HUMAN AND GUINEA-PIG LEUKOCYTE ADHESION GLYCOPROTEIN MO1 CHROMOSOMAL LOCALIZATION AND HOMOLOGY TO THE ALPHA SUBUNITS OF INTEGRINS
ARNAOUT M A; REMOLD-O'DONNELL E; PIERCE M W; HARRIS P; TENEN D G
RENAL AND GENETICS DIV., CHILDREN'S HOSP., BETH ISRAEL HOSP., BOSTON, MASS. 02115.
PROC NATL ACAD SCI U S A 85 (8). 1988. 2776-2780. CODEN: PNASA
Language: ENGLISH
The cell-surface glycoprotein Mo1 is a member of the family of leukocyte cell adhesion molecules (Leu-CAMs) that includes lymphocyte function-associated antigen 1 (LFA-1) and p150,95. Each Leu-CAM is a heterodimer with a distinct .alpha. subunit noncovalently associated with a common .beta. subunit. Leu-CAMs play crucial roles in cell-cell and cell-matrix interactions. We describe the isolation and analysis of two partial cDNA clones encoding the .alpha. subunit of the Leu-CAM Mo1 in humans and guinea pigs. A monoclonal antibody directed against an epitope in the carboxyl-terminal protion of the guinea pig .alpha. chain was used for immunoscreening a .lambda.gt11 expression library. The seqeunce of a 378-base-pair insert from one immunoreactive clone revealed a single continuous open reading frame encoding 126 amino acids including a 26-amino acid tryptic peptide isolated from the purified guinea pig .alpha. subunit

A cDNA clone of identical size was isolated from a human monocyte/lymphocyte cDNA library by using the guinea pig clone as a probe. The human clone also encoded a 126-amino acid peptide including the sequence of an additional tryptic peptide present in purified human Mo1.alpha. chain. RNA gel blots revealed that mature Mo1.alpha. chain mRNA is .apprxeq. 5 kilobaes in guinea pigs and humans. Southern analysis of DNA from hamster-human hybrids localized the human Mo1.alpha. chain to chromosome 16, which has been shown to contain the gene for the .alpha. chain of lymphocyte function-associated antigen 1. A comparison of the Mo1.alpha. chain coding region revealed significant homologies with carboxyl-terminal portions of the .alpha. subunits of fibronectin, vitronectin, and platelet IIb/IIIa receptors. These data suggest that the .alpha. subunit of Leu-CAMs evolved by gene duplication from a common ancestral gene and strengthen the hypothesis that the .alpha. subunits of these heterodimeric cell adhesion molecules on myeloid and lymphoid cells, platelets, and fibroblasts are evolutionarily related.

19/7/4 (Item 4 from file: 5)
0018184730 BIOSIS Number: 85086075
ANTIBODIES TO PEPTIDE DETERMINANTS IN TRANSFORMING GROWTH FACTOR BETA AND THEIR APPLICATIONS
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BIOCHEMISTRY 27 (2). 1988. 739-746. CODEN: BICHA
Language: ENGLISH
Polyclonal antibodies have been raised to a series of synthetic peptides which correspond to essentially all regions of the transforming growth factor .beta.1 (TGF-.beta.1) molecule. All antisera were evaluated for their abilities to react with TGF-.beta.1 and TGF-.beta.2 in either the native or reduced form in enzyme-linked immunosorbent assays, Western blots, and immunoprecipitation assays. While all antisera demonstrated some ability to recognize TGF-.beta.1 in these systems, there was limited cross-reactivity with TGF-.beta.2, suggesting that substantial sequence of conformational differences exist between the two growth factors. On Western blots 5-10 ng of purified human platelet TGF-.beta.1 could be detected when probed with affinity-purified peptide antisera generated against peptides corresponding to residues 48-77, 50-75, and 78-109 of the 112 amino acid TGF-.beta.1 monomer. Antisera raised against peptides 50-75 and 78-109 were most effective in immunoprecipitating reduced and native 125I-TGF-.beta.1, respectively. The antisera also were tested for their effectiveness in blocking the binding of 125I-TGF-.beta.1 to its receptor. Anti-peptide 78-109 and anti-peptide 50-75 blocked 80% and 40% of the binding, respectively, while antibodies against amino-terminal peptides were without effect. These data suggest that the carboxyl-terminal region of TGF-.beta.1 may play a significant role in the binding of the native ligand to its receptor.

19/7/5 (Item 5 from file: 5)
0018108847 BIOSIS Number: 85054316
COMPARISON OF COMPLEMENTARY DNA-DERIVED PROTEIN SEQUENCES OF THE HUMAN FIBRONECTIN AND VITRONECTIN RECEPTOR ALPHA-SUBUNITS AND PLATELET GLYCOPROTEIN IIB
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BIOCHEMISTRY 26 (25). 1987. 8158-8165. CODEN: BICHA
Language: ENGLISH
The fibronectin receptor (FnR), the vitronectin receptor (VnR), and the platelet membrane glycoprotein (GP) IIb-IIIa complex are members of a family of cell adhesion receptors, which consist of noncovalently associated .alpha.- and .beta.-subunits. The present study was designed to compare the cDNA-derived protein sequences of the .alpha.-subunits of human FnR, VnR, and platelet GP IIb. cDNA clones for the .alpha.-subunit of the FnR (FnR.alpha.) were obtained from a human umbilical vein endothelial (HUVE) cell library by using an oligonucleotide probe designed from a peptide sequence of platelet GP IIb. cDNA clones for platelet GP IIb were

isolated from a cDNA expression library of human erythroleukemia cells by using antibodies. cDNA clones of th[redacted]R .alpha.-subunit (VnR.alpha.) were obtained from the HUVE cell library by using an oligonucleotide probe from the partial cDNA sequence for the VnR.alpha.. Translation of these sequences showed that the FNR.alpha., the VnR.alpha., and GP IIb are composed of disulfide-linked large (858-871 amino acids) and small (137-158 amino acids) chains that are posttranslationally processed from a single mRNA. A single hydrophobic segment located near the carboxyl terminus of each small chain appears to be a transmembrane domain. The large chains appear to be entirely extracellular, and each contains four repeated putative Ca2+-binding domains of about 30 amino acids that have sequence similarities to other Ca2+-binding proteins. The identity among the protein sequences of the three receptor .alpha.-subunits ranges from 36.1% to 44.5%, with the Ca2+-binding domains having the greatest homology. These proteins apparently evolved by a process of gene duplication.

19/7/6 (Item 6 from file: 5)
0017518360 BIOSIS Number: 84006427
EXPRESSION OF MULTIPLE GROWTH FACTORS IN A HUMAN LUNG CANCER CELL LINE
BETHSHOLTZ C; BERGH J; BYWATER M; PETERSON M; JOHNSSON A; HELDIN C-H; OHLSSON R; KNOTT T J; SCOTT J; ET AL
DEP. OF PATHOL., UNIV. HOSP., S-751 85 UPPSALA, SWEDEN.
INT J CANCER 39 (4). 1987. 502-507. CODEN: IJCNA
Language: ENGLISH
U-1810, a human large-cell lung cancer line, was found to express a PDGF-like growth factor. 35S-cysteine labelling and immunoprecipitation revealed the synthesis and secretion of a 31-kDa PDGF-like protein. Serum-free conditioned medium contained PDGF-receptor-competing and mitogenic activity when tested on human fibroblasts. Whereas the receptor-competing activity was fully neutralized by anti-PDGF antibodies, the mitogenic activity was only partially affected. We therefore probed U-1810 mRNA with a panel of growth-factor DNA clones. We found expression of the genes for PDGF A- and B-chains, TGF-.alpha., TGF-.beta. and IGF-II but not EGF or IGF-I. U-1810 cells lacked specific binding sites for PDGF but showed specific binding of EGF and expressed EGF-receptor transcripts. Thus, U-1810 is an example of a human tumor cell line that expresses multiple growth factor genes; in the intact tumor the corresponding growth factors may operate in autocrine stimulation of the tumor cells as well as in paracrine growth reactions (i.e. stroma recruitment).

19/7/7 (Item 7 from file: 5)
0016649612 BIOSIS Number: 82070588
SYNTHESIS BY CULTURED HUMAN UMBILICAL VEIN ENDOTHELIAL CELLS OF TWO PROTEINS STRUCTURALLY AND IMMUNOLOGICALLY RELATED TO PLATELET MEMBRANE GLYCOPROTEINS IIB AND IIIA
NEWMAN P J; KAWAI Y; MONTGOMERY R R; KUNICKI T J
THE BLOOD CENTER OF SOUTHEASTERN WISCONSIN, MILWAUKEE, WISCONSIN 53233.
J CELL BIOL 103 (1). 1986. 81-86. CODEN: JCLBA
Language: ENGLISH
Human platelets participate in a number of adhesive interactions, including binding to exposed subendothelium after vascular injury, and platelet-platelet cohesion to form large aggregates. Platelet membrane glycoproteins (GP) IIb and IIIa constitute a receptor for fibrinogen that, together with fibrinogen and calcium, is largely responsible for mediating the formation of the primary hemostatic plug. Using highly specific polyclonal and monoclonal antibodies as probes, we could detect the presence of both of these glycoproteins in cultured human umbilical vein endothelial cells. Western-blot analysis showed that the endothelial cell analogues were similar in size to their platelet counterparts, and were present in cells that had been in culture for over 2 mo. Metabolic labeling of endothelium with [35S]methionine demonstrated that both GPIIb and GPIIIa were actively synthesized in cultures. Using the technique of crossed immunoelectrophoresis, evidence was obtained that the endothelial cell forms of GPIIb and GPIIIa may exist complexed to one another after solubilization in Triton X-100. The presence of GPIIb-IIIa analogues in cultured endothelial cells may provide an opportunity to examine the

structure, function, and synthesis of these two membrane glycoproteins, as well as provide a source of genetic material with which to begin detailed molecular genetic studies.

19/7/8 (Item 8 from file: 5)
0015713965 BIOSIS Number: 80103965
EXPRESSION OF THE C-SIS GENE AND SECRETION OF A PLATELET-DERIVED GROWTH FACTOR-LIKE PROTEIN BY SV-40-TRANSFORMED BHK CELLS
BETSHOLTZ C; BYWATER M; WESTERMARK B; BURK R R; HELDIN C-H
DEPARTMENT OF PATHOLOGY, UNIVERSITY HOSPITAL, S-751 85 UPPSALA, SWEDEN.
BIOCHEM BIOPHYS RES COMMUN 130 (2). 1985. 753-760. CODEN: BBRCA
Language: ENGLISH
SV40-transformed BHK cells express 2 transcripts, of 3.5 kb [kilobases] and 2.0 kb, that hybridized to human c-sis probe. Antibodies directed against human PDGF [platelet derived growth factor] specifically recognized a 31 kDa [kilodalton] protein in SV40/BHK cell conditioned medium, which upon reduction was split into 16 kDa species. Unfractioned conditioned medium and 1 of 2 growth factors isolated from SV40/BHK cells competed with 125I-PDGF for binding to its receptor. An SV40/BHK cell-derived growth factor is a hamster equivalent to human PDGF.

19/7/9 (Item 1 from file: 155)
06755924 89057924
Aberrant expression of receptors for platelet-derived growth factor in an anaplastic thyroid carcinoma cell line.
Heldin NE; Gustavsson B; Claesson-Welsh L; Hammacher A; Mark J; Heldin CH ; Westermark B
Department of Pathology, University of Uppsala, Sweden.
Proc Natl Acad Sci U S A (UNITED STATES) Dec 1988, 85 (23) p9302-6, ISSN 0027-8424 Journal Code: PV3
Languages: ENGLISH
Receptors for platelet-derived growth factor (PDGF) have previously only been found on cells of mesenchymal and glial origin. This study shows PDGF receptors on an anaplastic thyroid carcinoma cell line, C 643, that was found to express thyroglobulin mRNA, confirming its origin from thyroid epithelium. Northern blot analysis of poly(A)+ RNA hybridized with a human PDGF B-type receptor cDNA probe revealed a 5.4-kilobase transcript in the C 643 cells. The existence of receptor protein on the cell surface was shown by immunofluorescence microscopy with a PDGF receptor monoclonal antibody. Binding experiments with 125I-labeled dimeric forms of PDGF indicated that the cells contain B-, but not A-, type PDGF receptors. The addition of PDGF to C 643 membranes in the presence of [32P]ATP induced phosphorylation of the receptor. A polyclonal PDGF B-type receptor peptide antiserum was used to immunoprecipitate a receptor protein from metabolically labeled C 643 cells; the receptor was found to be 5-10 kDa larger than that in normal human fibroblasts. Removal of N-linked carbohydrates using endoglycosidase H resulted in deglycosylated receptor proteins of similar size in C 643 cells and fibroblasts, indicating differences in glycosylation patterns of the two receptor proteins. The aberrant expression of receptors might be crucial in tumor development by conferring a selective growth advantage to the cancer cells.

19/7/10 (Item 2 from file: 155)
06721594 89023594
Kidney epithelial cells express c-sis protooncogene and secrete PDGF-like protein.
Kartha S; Bradham DM; Grotendorst GR; Toback FG
Department of Medicine, University of Chicago, Illinois 60637.
Am J Physiol Oct 1988, 255 (4 Pt 2) pF800-6, ISSN 0002-9513 Journal Code: 3U8
Contract/Grant No.: DK-184313; DK-37227; DK-39689; +
Languages: ENGLISH
Nontransformed monkey kidney cells (BSC-1 line), used as a model for renal epithelium, were assayed for release of platelet-derived growth factor (PDGF)-like proteins. BSC-1 cells continuously released a mitogenic activity for fibroblasts and a chemoattractant activity for smooth muscle

cells, each of which was inhibited 80-90% by an antibody to human PDGF. A cDNA probe for the PDGF B-chain gene (c-sis), but not for the A-chain gene, hybridized to mRNA obtained from growing and quiescent cells. c-sis gene expression and PDGF-like protein secretion were studied in the presence of known growth-regulatory molecules. A secreted BSC-1 cell protein identical to transforming growth factor beta 2 inhibited DNA synthesis in growing cultures and induced marked accumulation of c-sis mRNA without a corresponding increase in the release of PDGF-like activity. Adenosine diphosphate stimulated DNA synthesis in quiescent cultures and enhanced both c-sis expression and release of PDGF-like activity. However, growing and quiescent cells did not express the PDGF receptor gene or exhibit a mitogenic response to authentic PDGF. Thus the PDGF-like protein released by these kidney epithelial cells could contribute to growth control by a paracrine mechanism.

19/7/11 (Item 3 from file: 155)
06706611 89008611

Role of IIb-IIIa-like glycoproteins in cell-substratum adhesion of human melanoma cells.

Knudsen KA; Smith L; Smith S; Karczewski J; Tuszynski GP
Lankenau Medical Research Center, Philadelphia, Pennsylvania 19151.

J Cell Physiol (UNITED STATES) Sep 1988, 136 (3) p471-8, ISSN 0021-9541 Journal Code: HNB
Contract/Grant No.: AR37945; HL28149
Languages: ENGLISH

The platelet fibrinogen receptor, glycoprotein complex IIb-IIIa, was isolated from human platelets by lectin and monoclonal antibody affinity chromatography and a polyclonal antiserum (anti-IIb-IIIa) was generated and used to probe for the presence and function of IIb-IIIa-like molecules in two adherent human cell lines. Both C32 melanoma cells and WI38 fibroblasts expressed a IIb-IIIa-like complex on their surface as indicated by immunoprecipitation of detergent extracts of surface radiolabeled cells. When added to cells plated in medium containing 10% serum, the anti-IIb-IIIa antiserum perturbed the adhesion of C32 melanoma cells, but not of WI38 fibroblasts. In a serum-free system, anti-IIb-IIIa antibodies inhibited attachment and spreading of C32 cells to fibrinogen, vitronectin, and fibronectin adsorbed to glass. Anti-IIb-IIIa had no effect on the attachment and spreading of WI38 cells to the extracellular matrix proteins, however. Thus, the IIb-IIIa-like complex appears to play a predominant role in cell-substratum adhesion of C32 cells, but not WI38 cells, and may result from the fact that, on a protein basis, the C32 melanoma cells express approximately 3 times more complex on their surface than do WI38 fibroblasts. The results suggest that the relative abundance of a particular adhesion receptor on the cell surface may govern its importance to cell-substratum adhesion.

19/7/12 (Item 4 from file: 155)
06693338 88338338

J774A.1 macrophage cell line produces PDGF-like and non-PDGF-like growth factors for bone cells.

Cheng SL; Rifas L; Shen V; Tong B; Pierce G; Deuel T; Peck WA
Department of Medicine Research, Jewish Hospital, Washington University Medical Center, St. Louis, MO 63110.

J Bone Miner Res Oct 1987, 2 (5) p467-74, ISSN 0884-0431 Journal Code: 130
Contract/Grant No.: AM 19855
Languages: ENGLISH

In light of evidence that macrophages participate in the local regulation of bone remodeling, we have examined the production of peptide stimulators of bone cell growth and specialization by the J774A.1 macrophage cell line. Cultured J774A.1 cells secrete growth-promoting activities which have an affinity for heparin. The first partially purified material, termed HEP I, appears to contain platelet-derived growth factor (PDGF)-like activity. It has a molecular weight of about 30,000 daltons, inhibits the binding of labeled PDGF to its receptors, reacts with polyclonal anti-human PDGF antibody, and exhibits mitogenic activity for osteoblasts, which is

partially blocked by anti-PDGF antisera. Like PDGF, HEP I is active in a wide variety of mesenchyme-derived cells, including osteoblasts, chondrocytes, smooth muscle cells, fibroblasts, 3T3 cells and NRK cells. The J774A.1 cells contain mRNA, which hybridizes to a v-sis DNA probe, suggesting that they express the c-sis gene, which contains the code for a PDGF-like protein. The second factor, HEP II, has an approximate molecular weight of 20,000 daltons and possesses substantial mitogenic activity for osteoblasts, chondrocytes, and smooth muscle cells, but is not mitogenic for fibroblasts, 3T3 cells, and NRK cells. HEP II appears to be a unique bone cell mitogen, which is distinct from the growth factors presently known. Neither HEP I nor HEP II contained interleukin 1, a macrophage product known to promote bone resorption and perhaps the growth and activity of osteoblasts.

19/7/13 (Item 5 from file: 155)
06572915 88217915

Cloning and expression of a cDNA coding for the human platelet-derived growth factor receptor: evidence for more than one receptor class.

Gronwald RG; Grant FJ; Haldeman BA; Hart CE; O'Hara PJ; Hagen FS; Ross R; Bowen-Pope DF; Murray MJ

Department of Pathology (SM-30), University of Washington, Seattle 98195.

Proc Natl Acad Sci U S A May 1988, 85 (10) p3435-9, ISSN 0027-8424
Journal Code: PV3
Contract/Grant No.: GM35501; HL18645; HL07312
Languages: ENGLISH

The complete nucleotide sequence of a cDNA encoding the human platelet-derived growth factor (PDGF) receptor is presented. The cDNA contains an open reading frame that codes for a protein of 1106 amino acids. Comparison to the mouse PDGF receptor reveals an overall amino acid sequence identity of 86%. This sequence identity rises to 98% in the cytoplasmic split tyrosine kinase domain. RNA blot hybridization analysis of poly(A)+ RNA from human dermal fibroblasts detects a major (approximately 5.7 kb) and a minor (approximately 4.8 kb) transcript using the cDNA as a probe. Baby hamster kidney cells, transfected with an expression vector containing the receptor cDNA, express an approximately equal to 190-kDa cell surface protein that is recognized by an anti-human PDGF receptor antibody. The recombinant PDGF receptor is functional in the transfected baby hamster kidney cells as demonstrated by ligand-induced phosphorylation of the receptor. Binding properties of the recombinant PDGF receptor were also assessed with pure preparations of BB and AB isoforms of PDGF (i.e., PDGF dimers composed of two B chains or an A and a B chain). Unlike human dermal fibroblasts, which bind both isoforms with high affinity, the transfected baby hamster kidney cells bind only the BB isoform of PDGF with high affinity. This observation is consistent with the existence of more than one PDGF receptor class.

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19/7/14 (Item 6 from file: 155)
06545151 88190151

Molecular cloning of the alpha subunit of human and guinea pig leukocyte adhesion glycoprotein Mo1: chromosomal localization and homology to the alpha subunits of integrins.

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Renal Division, Children's Hospital, Boston, MA.

Proc Natl Acad Sci U S A Apr 1988, 85 (8) p2776-80, ISSN 0027-8424
Journal Code: PV3
Contract/Grant No.: AI21963; CA21225; CA41456; +
Languages: ENGLISH

The cell-surface glycoprotein Mo1 is a member of the family of leukocyte cell adhesion molecules (Leu-CAMs) that includes lymphocyte function-associated antigen 1 (LFA-1) and p150,95. Each Leu-CAM is a heterodimer with a distinct alpha subunit noncovalently associated with a common beta subunit. Leu-CAMs play crucial roles in cell-cell and cell-matrix interactions. We describe the isolation and analysis of two partial cDNA clones encoding the alpha subunit of the Leu-CAM Mo1 in humans and guinea pigs. A monoclonal antibody directed against an epitope in the carboxyl-terminal portion of the guinea pig alpha chain was used for

immunoscreening a lambda gt11 expression library. The sequence of a 378-base-pair insert from one immunoreactive clone revealed a single continuous open reading frame encoding 126 amino acids including a 26-amino acid tryptic peptide isolated from the purified guinea pig alpha subunit. A cDNA clone of identical size was isolated from a human monocyte/lymphocyte cDNA library by using the guinea pig clone as a probe. The human clone also encoded a 126-amino acid peptide including the sequence of an additional tryptic peptide present in purified human Mo1 alpha chain. RNA gel blots revealed that mature Mo1 alpha chain mRNA is approximately 5 kilobases in guinea pigs and humans. Southern analysis of DNA from hamster-human hybrids localized the human Mo1 alpha chain to chromosome 16, which has been shown to contain the gene for the alpha chain of lymphocyte function-associated antigen 1. A comparison of the Mo1 alpha chain coding region revealed significant homologies with carboxyl-terminal portions of the alpha subunits of fibronectin, vitronectin, and platelet IIb/IIIa receptors. These data suggest that the alpha subunits of Leu-CAMs evolved by gene duplication from a common ancestral gene and strengthen the hypothesis that the alpha subunits of these heterodimeric cell adhesion molecules on myeloid and lymphoid cells, platelets, and fibroblasts are evolutionary related.

19/7/15 (Item 7 from file: 155)
06518661 88163661

Antibodies to peptide determinants in transforming growth factor beta and their applications.

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Biochemistry Jan 26 1988, 27 (2) p739-46, ISSN 0006-2960
Journal Code: A0G
Contract/Grant No.: 5F32AM07423
Languages: ENGLISH

Polyclonal antibodies have been raised to a series of synthetic peptides which correspond to essentially all regions of the transforming growth factor beta 1 (TGF-beta 1) molecule. All antisera were evaluated for their abilities to react with TGF-beta 1 and TGF-beta 2 in either the native or reduced form in enzyme-linked immunosorbent assays, Western blots, and immunoprecipitation assays. While all antisera demonstrated some ability to recognize TGF-beta 1 in these systems, there was limited cross-reactivity with TGF-beta 2, suggesting that substantial sequence or conformational differences exist between the two growth factors. On Western blots 5-10 ng of purified human platelet TGF-beta 1 could be detected when probed with affinity-purified peptide antisera generated against peptides corresponding to residues 48-77, 50-75, and 78-109 of the 112 amino acid TGF-beta 1 monomer. Antisera raised against peptides 50-75 and 78-109 were most effective in immunoprecipitating reduced and native 125I-TGF-beta 1, respectively. The antisera also were tested for their effectiveness in blocking the binding of 125I-TGF-beta 1 to its receptor. Anti-peptide 78-109 and anti-peptide 50-75 blocked 80% and 40% of the binding, respectively, while antibodies against amino-terminal peptides were without effect. These data suggest that the carboxyl-terminal region of TGF-beta 1 may play a significant role in the binding of the native ligand to its receptor.

19/7/16 (Item 8 from file: 155)
06518472 88163472

Comparison of cDNA-derived protein sequences of the human fibronectin and vitronectin receptor alpha-subunits and platelet glycoprotein IIb.

Fitzgerald LA; Poncz M; Steiner B; Rall SC Jr; Bennett JS; Phillips DR
Gladstone Foundation Laboratories for Cardiovascular Disease, San Francisco, California 94140-0608.

Biochemistry Dec 15 1987, 26 (25) p8158-65, ISSN 0006-2960
Journal Code: A0G
Contract/Grant No.: HL 28947; HL 32254
Languages: ENGLISH

The fibronectin receptor (FnR), the vitronectin receptor (VnR), and the

platelet membrane glycoprotein (GP) IIb-IIIa complex are members of a family of cell adhesion receptors, which consist of noncovalently associated alpha- and beta-subunits. The present study was designed to compare the cDNA-derived protein sequences of the alpha-subunits of human FnR, VnR, and platelet GP IIb. cDNA clones for the alpha-subunit of the FnR (FnR alpha) were obtained from a human umbilical vein endothelial (HUVE) cell library by using an oligonucleotide probe designed from a peptide sequence of platelet GP IIb. cDNA clones for platelet GP IIb were isolated from a cDNA expression library of human erythroleukemia cells by using antibodies. cDNA clones of the VnR alpha-subunit (VnR alpha) were obtained from the HUVE cell library by using an oligonucleotide probe from the partial cDNA sequence for the VnR alpha. Translation of these sequences showed that the FNR alpha, the VnR alpha, and GP IIb are composed of disulfide-linked large (858-871 amino acids) and small (137-158 amino acids) chains that are posttranslationally processed from a single mRNA. A single hydrophobic segment located near the carboxyl terminus of each small chain appears to be a transmembrane domain. The large chains appear to be entirely extracellular, and each contains four repeated putative Ca2+-binding domains of about 30 amino acids that have sequence similarities to other Ca2+-binding proteins. The identity among the protein sequences of the three receptor alpha-subunits ranges from 36.1% to 44.5%, with the Ca2+-binding domains having the greatest homology. These proteins apparently evolved by a process of gene duplication.

19/7/17 (Item 9 from file: 155)
06466521 88111521

Human proto-oncogene c-kit: a new cell surface receptor tyrosine kinase for an unidentified ligand.

Yarden Y; Kuang WJ; Yang-Feng T; Coussens L; Munemitsu S; Dull TJ; Chen E ; Schlessinger J; Francke U; Ullrich A

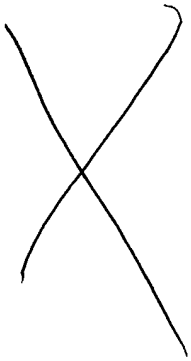
Department of Developmental Biology, Genentech, Inc., South San Francisco 94080.

EMBO J Nov 1987, 6 (11) p3341-51, ISSN 0261-4189 Journal Code: EMB

Contract/Grant No.: GM26105

Languages: ENGLISH

Structural features of v-kit, the oncogene of HZ4 feline sarcoma virus, suggested that this gene arose by transduction and truncation of cellular sequences. Complementary DNA cloning of the human proto-oncogene coding for a receptor tyrosine kinase confirmed this possibility: c-kit encodes a transmembrane glycoprotein that is structurally related to the receptor for macrophage growth factor (CSF-1) and the receptor for platelet-derived growth factor. The c-kit gene is widely expressed as a single, 5-kb transcript, and it is localized to human chromosome 4 and to mouse chromosome 5. A c-kit peptide antibody permitted the identification of a 145,000 dalton c-kit gene product that is inserted in the cellular plasma membrane and is capable of self-phosphorylation on tyrosine residues in both human glioblastoma cells and transfected mouse fibroblasts. Our results suggest that p145c-kit functions as a cell surface receptor for an as yet unidentified ligand. Furthermore, carboxy- and amino-terminal truncations that occurred during the viral transduction process are likely to have generated the transformation potential of v-kit.



19/7/18 (Item 10 from file: 155)
06446177 88091177

Expression of genes for platelet-derived growth factor in adult human venous endothelium. A possible non-platelet-dependent cause of intimal hyperplasia in vein grafts and perianastomotic areas of vascular prostheses.

Limanni A; Fleming T; Molina R; Hufnagel H; Cunningham RE; Cruess DF; Sharefkin JB

Department of Surgery, Uniformed Services University of the Health Sciences, Bethesda, MD 20814.

J Vasc Surg Jan 1988, 7 (1) p10-20, ISSN 0741-5214 Journal Code: KD2

Contract/Grant No.: R01 HL33931

Languages: ENGLISH

Neointimal fibromuscular hyperplasia (NFH) in vein grafts and perianastomotic zones of vascular anastheses has been attributed to the effects of platelet-derived growth factor (PDGF) released by platelets interacting with bypass conduits. But inhibition of platelet aggregation often fails to prevent NFH, and recurrent growth of intact, platelet-free endothelium over perianastomotic areas where NFH occurs is inconsistent with the concept of sustained PDGF release from platelets causing NFH progression at late times after surgical procedures. Cultured bovine aortic endothelial cells (ECs) and human umbilical vein ECs have been shown to release a PDGF-like molecule. We report that confluent cultured fourth passage adult human saphenous vein ECs (AHSVECs) grown in the presence of heparin (100 micrograms/ml) and retina-derived growth factor (RDGF) studied by Northern blotting transcribed a messenger ribonucleic acid (mRNA) of 3.9 kb, strongly hybridizing to PDGF B chain probes, and two species of 2.0 and 2.6 kb hybridizing to PDGF A chain probes. Withdrawal of RDGF and heparin from these cultures for 48 hours before mRNA extraction amplified the scanning densitometric mRNA signal per cell by 8.0 +/- 7.6 fold (mean +/- SD) (N = 4 cultures) for B chain mRNA and 5.2 +/- 3.6 fold (N = 3 cultures) for A chain mRNA. In addition, AHSVEC cultures released a PDGF-like substance, because 50% vol/vol AHSVEC-conditioned serum-free medium increased tritiated thymidine uptake elevenfold in PDGF receptor-bearing 3T3 cells whereas an excess (50 micrograms/ml) of nonspecific goat anti-human-PDGF antibody significantly reduced this increase by a mean of 30% to 7.0 +/- 3.4 fold (N = 6 trials, p less than 0.001). Flow cytometry determined AHSVEC cultures to be proliferating with a mean of 6.2% +/- 1.9% (N = 3 culture lines) of ECs in S phase even at confluence when deprived of EC mitogens for 48 hours. Adult human ECs, which proliferate on bypass conduits and host vessels after perioperative injury, may play a role in causing NFH by stimulating proliferation of adjacent smooth muscle cells. Prevention of NFH may require not only antiplatelet agents but also ways to prevent EC release of smooth muscle cell mitogens in response to perioperative EC injury.

19/7/19 (Item 11 from file: 155)
06190630 87164630

Expression of multiple growth factors in a human lung cancer cell line.

Betsholtz C; Bergh J; Bywater M; Pettersson M; Johnsson A; Heldin CH; Ohlsson R; Knott TJ; Scott J; Bell GI; et al

Int J Cancer Apr 15 1987, 39 (4) p502-7, ISSN 0020-7136
Journal Code: G0U
Languages: ENGLISH

U-1810, a human large-cell lung cancer line, was found to express a PDGF-like growth factor. 35S-cysteine labelling and immunoprecipitation revealed the synthesis and secretion of a 31-kDa PDGF-like protein. Serum-free conditioned medium contained PDGF-receptor-competing and mitogenic activity when tested on human fibroblasts. Whereas the receptor-competing activity was fully neutralized by anti-PDGF antibodies, the mitogenic activity was only partially affected. We therefore probed U-1810 mRNA with a panel of growth-factor DNA clones. We found expression of the genes for PDGF A- and B-chains, TGF-alpha, TGF-beta and IGF-II but not EGF or IGF-I. U-1810 cells lacked specific binding sites for PDGF but showed specific binding of EGF and expressed EGF-receptor transcripts. Thus, U-1810 is an example of a human tumor cell line that expresses multiple growth factor genes; in the intact tumor the corresponding growth factors may operate in autocrine stimulation of the tumor cells as well as in paracrine growth reactions (i.e. stroma recruitment).

19/7/20 (Item 12 from file: 155)
06183592 87157592

Partial amino acid sequence of human thrombospondin as determined by analysis of cDNA clones: homology to malarial circumsporozoite proteins.

Kobayashi S; Eden-McCutchan F; Framson P; Bornstein P

Biochemistry Dec 30 1986, 25 (26) p8418-25, ISSN 0006-2960
Journal Code: A0G
Contract/Grant No.: AM 11248; HL 18645; DE 02600
Languages: ENGLISH

A lambda gt 11 library prepared from human umbilical vein endothelial cell RNA was screened for cDNAs encoding thrombospondin. Reagents included a monospecific antibody to human thrombospondin and a mixture of four synthetic oligodeoxyribonucleotides derived from an amino acid sequence near the NH2 terminus of mature human thrombospondin. Two series of cDNA clones coding for sequences at the 5′ and 3′ ends of thrombospondin mRNA, respectively, were isolated. The nucleotide sequence of a 1.3-kilobase (kb) 5′ clone (lambda TS-33) coded for 99 bases of 5′ untranslated RNA, a signal peptide of 18 amino acids, and the first 379 amino acids of thrombospondin. Northern blot analysis with lambda TS-33 detected a single mRNA species of approximately 6.0 kb in rat aortic smooth muscle cell RNA. Thrombospondin mRNA levels increased rapidly, but transiently, in quiescent smooth muscle cells treated with platelet-derived growth factor. The kinetics of this response were very similar to those of the thrombospondin protein to this growth factor. There was significant homology in amino acid sequence between thrombospondin and a conserved region in the circumsporozoite protein of two malarial sporozoites. This region of thrombospondin may therefore represent a potential recognition site for a cell surface thrombospondin receptor.

19/7/21 (Item 13 from file: 155)
06042914 87016914

Human melanoma cell lines of primary and metastatic origin express the genes encoding the chains of platelet-derived growth factor (PDGF) and produce a PDGF-like growth factor.

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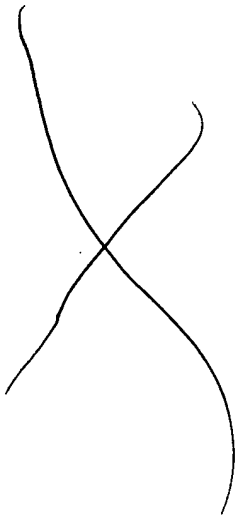
Proc Natl Acad Sci U S A Oct 1986, 83 (19) p7197-200, ISSN 0027-8424 Journal Code: PV3

Contract/Grant No.: CA-25874; CA-21124; CA-10815

Languages: ENGLISH

Normal human melanocytes and five human melanoma cell lines were analyzed for production of platelet-derived growth factor (PDGF)-like activity. Three of the melanoma cell lines released an activity that inhibited binding of 125I-labeled PDGF to human foreskin fibroblasts and stimulated [3H]thymidine incorporation in such cells. These activities were inhibited by the addition of anti-PDGF antibodies. All three factor-producing cell lines were derived from the same patient--one originated from the primary tumor (WM 115), and two were from individual lymph-node metastases (WM 239A and WM 266-4). The factor produced by WM 266-4 cells was characterized biochemically in detail. Immunoprecipitated, the metabolically labeled factor migrated in NaDod-SO4/gel electrophoresis as a homogeneous Mr 31,000 species, which under reducing conditions was resolved into two species of Mr 16,500 and Mr 17,000, implying a dimeric structure of the molecule. The factor was purified to homogeneity. Analysis by reverse-phase high-pressure liquid chromatography of reduced and alkylated factor revealed an elution pattern identical to that of PDGF A chains. Thus, the native molecule appears to be a homodimer of PDGF A chains. Blot-hybridization analysis of poly(A)+ RNA from the cell lines with 32P-labeled PDGF A chain and B chain (SIS product) cDNA probes revealed a relative abundance of B chain transcripts in the cell line originating from the primary tumor tissue only but expression of A chain in all three cell lines. We conclude that the two structural genes encoding each of the subunit chains of PDGF can be expressed in human melanoma cells and that the two genes can be independently expressed in such cells.



19/7/22 (Item 14 from file: 155)
05950036 86251036

Synthesis by cultured human umbilical vein endothelial cells of two proteins structurally and immunologically related to platelet membrane glycoproteins IIb and IIIa.

Newman PJ; Kawai Y; Montgomery RR; Kunicki TJ

J Cell Biol Jul 1986, 103 (1) [redacted], ISSN 0021-9525 Journal Code: HMV

Contract/Grant No.: HL-32279, HL-33721, HL-28444, +

Languages: ENGLISH

Human platelets participate in [redacted]umber of adhesive interactions, including binding to exposed subendothelium after vascular injury, and platelet-platelet cohesion to form large aggregates. Platelet membrane glycoproteins (GP) IIb and IIIa constitute a receptor for fibrinogen that, together with fibrinogen and calcium, is largely responsible for mediating the formation of the primary hemostatic plug. Using highly specific polyclonal and monoclonal antibodies as probes, we could detect the presence of both of these glycoproteins in cultured human umbilical vein endothelial cells. Western-blot analysis showed that the endothelial cell analogues were similar in size to their platelet counterparts, and were present in cells that had been in culture for over 2 mo. Metabolic labeling of endothelium with [35S]methionine demonstrated that both GPIIb and GPIIIa were actively synthesized in culture. Using the technique of crossed immunoelectrophoresis, evidence was obtained that the endothelial cell forms of GPIIb and GPIIIa may exist complexed to one another after solubilization in Triton X-100. The presence of GPIIb-IIIa analogues in cultured endothelial cells may provide an opportunity to examine the structure, function, and synthesis of these two membrane glycoproteins, as well as provide a source of genetic material with which to begin detailed molecular genetic studies.

19/7/23 (Item 15 from file: 155)
05861242 86162242

Paracrine action of transforming growth factors.

Gol-Winkler R

Clin Endocrinol Metab Feb 1986, 15 (1) p99-115, ISSN 0300-595X
Journal Code: DCR

Languages: ENGLISH

Document Type: Review

Polypeptide growth factors form a class of regulatory molecules which exert their effects by binding to specific receptors present on the cell surface. Most of the time the exact role of these factors in the healthy body is unknown. Some, like PDGF and TGF beta, seem to be involved in wound healing. Others, like EGF, promote epithelial cell growth and differentiation. The site of synthesis of most polypeptide growth factors is unknown. Their target can be identified by detecting the cells which present the specific receptors at their surface. It is though that polypeptide growth factors have a paracrine mode of action. Many different cancerous cells produce polypeptide growth factors and the appropriate receptors. Thus, they are able to stimulate their own growth in an autocrine fashion. Recently, some polypeptide growth factors and receptor genes or cDNAs have been molecularly cloned. Growth factor genes and messengers are much more complex than would be expected from the size of the polypeptide. Some cDNAs have been introduced into bacterial expression vectors and large amounts of the factors have been produced by bacteria. New tools, such as molecular probes and specific antibodies, are thus now available to investigate the production of the growth factors and their receptors. The same tools will facilitate the identification and understanding of the molecular mechanism whereby cancerous cells produce the growth factors and the appropriate receptors simultaneously. The importance of growth factors and receptors in cancer is stressed by the finding that three oncogenes are in fact the genes coding for one growth factor and two receptors. Finally, the molecular probes and the specific antibodies raised against these molecules can be used to identify precisely the growth factor(s) and receptor(s) produced abnormally in cancers. Antibodies that inhibit specifically the interaction of this very growth factor with its receptor could then be developed, thus allowing human tumour cell growth to be controlled. (117 Refs.)

19/7/24 (Item 16 from file: 155)
05769233 86070233

Determinants of von Willebrand factor activity elicited by ristocetin and botrocetin: studies on a human von Willebrand factor-binding antibody.

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Semin Thromb Hemost Oct 1985, 11 (4) p337-41, ISSN 0094-6176
Journal Code: UKS
Contract/Grant No.: HL-01648
Languages: ENGLISH

The three main probes for functional vWF activity--ristocetin, botrocetin, and the PAggF test--and similarities and differences in their elicited vWF activities have been reviewed. Emphasis has been placed on the technologies dependent on these probes, with a brief description of a series of relatively simple and sensitive tests developed in this laboratory. These tests include the development of the PAF test for vWF in certain animal plasmas; the development and use of fixed lyophilized platelets that retain receptor activity for vWF; the purification of botrocetin (venom coagglutinin) freed of thrombinlike enzymes and its use in vWF assays; the development of macroscopic platelet aggregation tests for screening and assay of vWF; and the application of the macroscopic test for rapid screening and quantitation of human plasmas for acquired inhibitors of vWF utilizing each of the three probes. Historically, the similarities of the ristocetin and botrocetin probes were first observed. For normal human plasmas and for patients with classic vWD, both homozygous or heterozygous, similar values for vWF were obtained with these two probes. Similar platelet binding of vWF in the presence of the two probes was likewise noted. However, further studies of these two probes revealed striking differences. Especially important for study of animal plasmas generally as well as a canine model of vWD was the observation that the vWF in all animal plasmas tested with botrocetin was highly reactive, whereas with ristocetin nearly all plasmas were resistant. Similarly, all animal platelets tested for vWF-dependent aggregation with the two probes were highly reactive with botrocetin, but inactive with ristocetin.(ABSTRACT TRUNCATED AT 250 WORDS)

19/7/25 (Item 17 from file: 155)
05663463 85279463

Expression of the c-sis gene and secretion of a platelet-derived growth factor-like protein by simian virus 40-transformed BHK cells.

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Biochem Biophys Res Commun Jul 31 1985, 130 (2) p753-60, ISSN 0006-291X Journal Code: 9Y8

Languages: ENGLISH

SV40-transformed BHK cells were shown to express two transcripts, of 3.5 kb and 2.0 kb, that hybridised to a human c-sis probe. Antibodies directed against human PDGF specifically recognized a 31 kDa protein in SV40/BHK cell conditioned medium, which upon reduction was split into 16 kDa species. Unfractionated conditioned medium and one of two growth factors isolated from SV40/BHK cells competed with 125I-PDGF for binding to its receptor. The present communication thus provides compelling evidence that an SV40/BHK cell-derived growth factor is a hamster equivalent to human PDGF.

19/7/26 (Item 18 from file: 155)
05458453 85074453

Coexpression of a PDGF-like growth factor and PDGF receptors in a human osteosarcoma cell line: implications for autocrine receptor activation.

Betsholtz C; Westermark B; Ek B; Heldin CH

Cell Dec 1984, 39 (3 Pt 2) p447-57, ISSN 0092-8674 Journal Code: CQ4

Languages: ENGLISH

The expression of both a PDGF-like growth factor and functional PDGF receptors within a clonal human osteosarcoma cell line (U-2 OS Cl 6) is demonstrated. These molecules are able to interact and induce tyrosine-specific phosphorylation and early actin reorganization in the osteosarcoma cells, effects similar to those that PDGF induces in normal responsive cells. Furthermore, immunoprecipitation with an antiserum against phosphotyrosine revealed th[redacted] 115 kd protein was constitutively phosphorylated in U-2 OS Cl 6 cell[redacted] phosphorylated protein of similar apparent molecular weight has been found in human fibroblasts, but only

after stimulation with PDGF. These data indicate that the PDGF-receptor-dependent pathway is constitutively activated in this cell line. Extracellularly added PDGF antibodies did not, however, affect the transformed properties or growth rate of U-2 OS Cl 6 cells in vitro. This indicates that autocrine PDGF receptor activation may be insignificant for maintaining the transformed state of this tumor cell line, or that autocrine receptor activation occurs in a compartment where it is inaccessible to extracellularly added antibodies.

19/7/27 (Item 1 from file: 357)
077484 DBA Accession No.: 88-08333
Cloning and expression of a cDNA coding for the human platelet-derived growth factor receptor: evidence for more than one receptor class - DNA sequence determination
AUTHOR: Gronwald R G K; Grant F J; Haldeman B A; Hart C E; O'Hara P J; +Bowen-Pope D F
CORPORATE AFFILIATE: Zymogenetics
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JOURNAL: Proc.Natl.Acad.Sci.U.S.A. (85, 10, 3435-39) CODEN: PNASA6
PUBLICATION YEAR: 1988 LANGUAGE: English
ABSTRACT: About 1 million phage from a random-primed human dermal fibroblast (SK5) cDNA library were screened with a mixture of 3 oligonucleotide probes corresponding to sequences of mouse platelet-derived growth factor (PDGR) receptor. 2 Clones, RP41 and RP51, corresponding to the 5' end of the cDNA were isolated. Further screening of the library using a 630 bp SstI/EcoRI fragment derived from the 3' end of clone RP41 yielded clone OT91, which contained the 3' end coding region. Clones RP51, RP41 and OP91 were ligated to yield a cDNA which was subcloned into vector plasmid pDX, and the cDNA sequence was determined. The cDNA contained an open reading frame encoding a protein of 1106 amino acids. There was 86% amino acid homology with the mouse PDGF receptor. BHK cells transfected with phPDGFR produced an approximately 190-kDa cell surface protein recognized by an anti-human PDGR receptor antibody. The transfectants expressed a high-affinity receptor specific for the BB form of PDGF. (40 ref)
?display sets

Set	Items	Description
S1	2433	AU=MATSUI, T? OR AU=MATSUI T?
S2	1107	AU=AARONSON, S? OR AU=AARONSON S?
S3	1844	AU=PIERCE, J? OR AU=PIERCE J?
S4	5292	S1 OR S2 OR S3
S5	150249	PDGF? OR PLATELET?
S6	147597	S5 NOT PY=1990
S7	118	S6 AND S4
S8	11567	RECEPTOR? AND S6
S9	35	S8 AND S4
S10	14	S9 NOT PY=1989
S11	2822127	GENE? OR SEQUENCE?
S12	134510	S6 NOT PY=1989
S13	134496	S12 NOT S9
S14	9509	S13 AND RECEPTOR?
S15	1164	S14 AND S11
S16	209507	PROBE? OR HYBRIDIZ?
S17	82	S15 AND S16
S18	585675	ANTIBOD?
S19	27	S17 AND S18